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¢%^STN;HighlightOn=  ***;HighlightOff=***  ;
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 NEWS  1            Web Page for STN Seminar Schedule - N. America
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 NEWS  3   JAN 16   CAS patent coverage enhanced to include exemplified
                    prophetic substances
 NEWS  4   JAN 28   USPATFULL, USPAT2, and USPATOLD enhanced with new
                    custom IPC display formats
 NEWS  5   JAN 28   MARPAT searching enhanced
 NEWS  6   JAN 28   USGENE now provides USPTO sequence data within 3 days
                    of publication
 NEWS  7   JAN 28   TOXCENTER enhanced with reloaded MEDLINE segment
 NEWS  8   JAN 28   MEDLINE and LMEDLINE reloaded with enhancements
 NEWS  9   FEB 08   STN Express, Version 8.3, now available
 NEWS 10   FEB 20   PCI now available as a replacement to DPCI
 NEWS 11   FEB 25   IFIREF reloaded with enhancements
 NEWS 12   FEB 25   IMSPRODUCT reloaded with enhancements
 NEWS 13   FEB 29   WPINDEX/WPIDS/WPIX enhanced with ECLA and current
                    U.S. National Patent Classification
 NEWS 14   MAR 31   IFICDB, IFIPAT, and IFIUDB enhanced with new custom
                    IPC display formats
 NEWS 15   MAR 31   CAS REGISTRY enhanced with additional experimental
                    spectra
 NEWS 16   MAR 31   CA/CAplus and CASREACT patent number format for U.S.
                    applications updated
 NEWS 17   MAR 31   LPCI now available as a replacement to LDPCI
 NEWS 18   MAR 31   EMBASE, EMBAL, and LEMBASE reloaded with enhancements
 NEWS 19   APR 04   STN AnaVist, Version 1, to be discontinued
 NEWS 20   APR 15   WPIDS, WPINDEX, and WPIX enhanced with new
                    predefined hit display formats
 NEWS 21   APR 28   EMBASE Controlled Term thesaurus enhanced
 NEWS 22   APR 28   IMSRESEARCH reloaded with enhancements

 NEWS EXPRESS  FEBRUARY 08 CURRENT WINDOWS VERSION IS V8.3,
              AND CURRENT DISCOVER FILE IS DATED 20 FEBRUARY 2008

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* * * * * * * * * * * * * * STN  Columbus  * * * * * * * * * * * * * * * *

FILE 'HOME' ENTERED AT 17:51:25 ON 26 MAY 2008

=> microfluidic and red blood cells
MICROFLUIDIC IS NOT A RECOGNIZED COMMAND
The previous command name entered was not recognized by the system.
For a list of commands available to you in the current file, enter
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"HELP COMMANDS" at an arrow prompt (=>).

=> s microfluidic and red blood cells
THIS COMMAND NOT AVAILABLE IN THE CURRENT FILE
Some commands only work in certain files. For example, the EXPAND
command can only be used to look at the index in a file which has an
index. Enter "HELP COMMANDS" at an arrow prompt (=>) for a list of
commands which can be used in this file.

=> index bioscience
FILE 'DRUGMONOG' ACCESS NOT AUTHORIZED
COST IN U.S. DOLLARS                               SINCE FILE      TOTAL
                                                        ENTRY    SESSION
FULL ESTIMATED COST                                      0.42       0.42

INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, ANTE, AQUALINE,
       AQUASCI, BIOENG, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS,
       CEABA-VTB, CIN, CONFSCI, CROPB, CROPU, DDFB, DDFU, DGENE, DISSABS, DRUGB,
       DRUGMONOG2, DRUGU, EMBAL, EMBASE, ...' ENTERED AT 17:52:32 ON 26 MAY 2008
69 FILES IN THE FILE LIST IN STNINDEX

Enter SET DETAIL ON to see search term postings or to view
search error messages that display as 0* with SET DETAIL OFF.

=> s microfluidic and red blood cells
          8    FILE ANABSTR
          1    FILE ANTE
          8    FILE BIOENG
         28    FILE BIOSIS
          1    FILE BIOTECHABS
          1    FILE BIOTECHDS
          1    FILE BIOTECHNO
          2    FILE CABA
         57    FILE CAPLUS
          1    FILE CEABA-VTB
  22 FILES SEARCHED...
          7    FILE DISSABS
         21    FILE EMBASE
         25    FILE ESBIOBASE
         65    FILE IFIPAT
          8    FILE LIFESCI
         27    FILE MEDLINE
          1    FILE NTIS
         15    FILE PASCAL
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          1    FILE PHIN
          8    FILE PROMT
         66    FILE SCISEARCH
          4    FILE TOXCENTER
        580    FILE USPATFULL
        113    FILE USPAT2
         25    FILE WPIDS
         25    FILE WPINDEX
  26 FILES HAVE ONE OR MORE ANSWERS,   69 FILES SEARCHED IN STNINDEX
L1    QUE MICROFLUIDIC AND RED BLOOD CELLS

=> s cell(p)size and cell(p)bind? and L1
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         0*    FILE ANTE
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         0*    FILE BIOENG
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         0*    FILE BIOTECHNO
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         0*    FILE CIN
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          0*    FILE EOREGE
          0*    FILE EROSTI
          0*    FILE ESTA
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          1     FILE MEDLINE
          0*    FILE NTIS
          0*    FILE NUTRACEUT
          0*    FILE PASCAL
          0*    FILE PHARMAML
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        255     FILE USPATFULL
         48     FILE USPAT2
          0*    FILE WATER
          2     FILE WPIDS
          3     FILE WPINDEX
   7 FILES HAVE ONE OR MORE ANSWERS,   69 FILES SEARCHED IN STNINDEX
L2    QUE CELL(P) SIZE AND CELL(P) BIND? AND L1
=> file caplus ifipat medline uspatfull uspat2
COST IN U.S. DOLLARS                                   SINCE FILE      TOTAL
                                                            ENTRY    SESSION
FULL ESTIMATED COST                                          4.55       4.97
FILE 'CAPLUS' ENTERED AT 17:56:56 ON 26 MAY 2008
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FILE 'MEDLINE' ENTERED AT 17:56:56 ON 26 MAY 2008
FILE 'USPATFULL' ENTERED AT 17:56:56 ON 26 MAY 2008
CA INDEXING COPYRIGHT (C) 2008 AMERICAN CHEMICAL SOCIETY (ACS)
FILE 'USPAT2' ENTERED AT 17:56:56 ON 26 MAY 2008
CA INDEXING COPYRIGHT (C) 2008 AMERICAN CHEMICAL SOCIETY (ACS)
=> s l2
L3          325 L2
=> dup rem l3
PROCESSING COMPLETED FOR L3
L4            259 DUP REM L3 (66 DUPLICATES REMOVED)
=> s l4 and enrich?
L5          163 L4 AND ENRICH?
=> s l5 and hist
L6            0 L5 AND HIST
=> s l5 and bind?
L7          163 L5 AND BIND?
=> s l7 and obstacle?
L8           70 L7 AND OBSTACLE?
=> s l8 and binding to obstacle?
L9            0 L8 AND BINDING TO OBSTACLE?
=> s l8 and microfluidic device
L10          19 L8 AND MICROFLUIDIC DEVICE
=> d l10 1-19
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L10 ANSWER 1 OF 19 CAPLUS COPYRIGHT 2008 ACS on STN
AN 2004:282081 CAPLUS <<LOGINID::20080526>>
DN 140:283837
TI ***Microfluidic*** ***device*** for cell separation and uses thereof
IN Toner, Mehmet; Truskey, George; Kapur, Ravi
PA The General Hospital Corporation, USA; GPB Scientific LLC
SO PCT Int. Appl., 81 pp.
CODEN: PIXXD2
DT Patent
LA English
FAN.CNT 1

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
PI	WO 2004029221	A2	20040408	WO 2003-US30965	20030929
	WO 2004029221	A3	20040513		

W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NI, NO, NZ, OM, PG, PH, PL, PT, RO, RU, SC, SD, SE, SG, SK, SL, SY, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, YU, ZA, ZM, ZW
RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZM, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HU, IE, IT, LU, MC, NL, PT, RO, SE, SI, SK, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, ML, MR, NE, SN, TD, TG

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
	CA 2500392	A1	20040408	CA 2003-2500392	20030929
	AU 2003277153	A1	20040419	AU 2003-277153	20030929
	EP 1569510	A2	20050907	EP 2003-798902	20030929

R: AT, BE, CH, DE, DK, ES, FR, GB, GR, IT, LI, LU, NL, SE, MC, PT, IE, SI, LT, LV, FI, RO, MK, CY, AL, TR, BG, CZ, EE, HU, SK

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
	JP 2006501449	T	20060112	JP 2004-540301	20030929
	US 20060134599	A1	20060622	US 2005-529453	20051219
	US 20070172903	A1	20070726	US 2007-726230	20070321
	US 20070231851	A1	20071004	US 2007-726276	20070321
	US 20070259424	A1	20071108	US 2007-726231	20070321
	US 20070264675	A1	20071115	US 2007-800940	20070508
PRAI	US 2002-414065P	P	20020927		
	US 2002-414102P	P	20020927		
	US 2002-414258P	P	20020927		
	WO 2003-US30965	W	20030929		
	US 2005-529453	A3	20051219		

L10 ANSWER 2 OF 19 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11614553 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 2007264675 A1 20071115
AI US 2007-800940 20070508
RLI US 2005-529453 20051219 CONTINUATION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
FI US 2007264675 20071115
DT Utility; Patent Application - First Publication
FS CHEMICAL
APPLICATION
ED Entered STN: 16 Nov 2007
Last Updated on STN: 13 Dec 2007
CLMN 27
GI 25 Figure(s).
FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.
FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.
FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.

FIG. 4 is an illustration of the outlet channels of the device.

FIG. 5 is an illustration of a device for ***cell*** lysis.

FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.

FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.

FIG. 8 is an exploded view of a ***cell*** ***binding*** device.

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.

FIG. 10 is an illustration of types of ***obstacles***.

FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.

FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.

FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.3391. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 99%.

FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.

FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.

FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.

FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.

FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.

FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.

FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.

FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.

FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.

FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***

L10 ANSWER 3 OF 19 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11609304 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 2007259424 A1 20071108
AI US 2007-726231 20070321
RLI US 2005-529453 20051219 DIVISION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
FI US 2007259424 20071108
DT Utility; Patent Application - First Publication
FS CHEMICAL APPLICATION
ED Entered STN: 14 Nov 2007
Last Updated on STN: 13 Dec 2007
CLMN 34
GI 25 Figure(s).

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.
FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.
FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.
FIG. 4 is an illustration of the outlet channels of the device.
FIG. 5 is an illustration of a device for ***cell*** lysis.
FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.
FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.
FIG. 8 is an exploded view of a ***cell*** ***binding*** device.
FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.
FIG. 10 is an illustration of types of ***obstacles***.
FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.
FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.
FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.
FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.
FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.3391. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 93%.
FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer

cells, normal connective tissue cells, or cells from cord blood into
defined number of cells from buffy coat of adult blood. The ratio of the
contaminating cells to target cells was incrementally increased 5 log
with as few as 10 target cells in the mixture. Yield was computed as the
difference between number of spiked target cells captured on posts and
number of cells spiked into the sample.
FIG. 17 is an illustration of various views of the inlet and outlets of a
cell ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell***
binding device.
FIG. 19 is an illustration of a mixture of cells flowing through a
cell ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device
for trapping different types of cells in series. FIG. 20B is an
illustration of a ***cell*** ***binding*** device for trapping
different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device
that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood***
cells adhered to an ***obstacle*** of the invention. FIG. 22B
is a fluorescent micrograph showing the results of a FISH analysis of a
fetal red blood ***cell*** attached to an ***obstacle*** of the
invention. FIG. 22C is a close up micrograph of FIG. 22B showing the
individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in
which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based
separation. FIG. 24B is an electron micrograph of a device for
size based separation.
FIG. 25 is a schematic representation of a device of the invention for
isolating and analyzing fetal ***red*** ***blood*** ***cells***
.

L10 ANSWER 4 OF 19 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11581736 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES
THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 2007231851 A1 20071004
AI US 2007-726276 20070321
RLI US 2005-529453 20051219 CONTINUATION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2003-414258P 20020927 (Provisional)
FI US 2007231851 20071004
DT Utility; Patent Application - First Publication
FS CHEMICAL
APPLICATION
ED Entered STN: 8 Oct 2007
Last Updated on STN: 9 Nov 2007
CLMN 33
GI 25 Figure(s).
FIG. 1 is a schematic layout of a ***microfluidic*** ***device***
that enables selective lysis of cells.
FIG. 2 is an illustration of the channel layout for the introduction of
three fluids to the device, e.g., blood sample, lysis buffer, and
diluent.
FIG. 3 is an illustration of a repeating unit of the reaction chamber of
the device where a sample of cells is passively mixed with a lysis
buffer. In one example, 133 units are connected to form the reaction
chamber.
FIG. 4 is an illustration of the outlet channels of the device.
FIG. 5 is an illustration of a device for ***cell*** lysis.
FIGS. 6A and 6B are illustrations of a method for the fabrication of a
device of the invention.
FIG. 7 is a schematic diagram of a ***cell*** ***binding***
device.
FIG. 8 is an exploded view of a ***cell*** ***binding*** device.
FIG. 9 is an illustration of ***obstacles*** in a ***cell***
binding device.
FIG. 10 is an illustration of types of ***obstacles*** .

FIG. 11A is a schematic representation of a square array of ***obstacle*** . The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacle*** . The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array. FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.

FIG. 15 is a schematic representation of the arrangement of ***obstacle*** for higher efficiency capture for an equilateral triangular array of ***obstacle*** in a staggered array. The capture radius rcap2=0.329 l. The ***obstacle*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 99%.

FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.

FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.

FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.

FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.

FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.

FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.

FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell*** .

FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.

FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.

FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells*** .

Figures are not necessarily to scale.

AN 11522821 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 2007172903 A1 20070726
AI US 2007-726230 20070321

RLI US 2005-528453 20051219 DIVISION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
PI US 2007172903 20070726
DT Utility; Patent Application - First Publication
FS CHEMICAL APPLICATION
ED Entered STN: 26 Jul 2007
Last Updated on STN: 16 Aug 2007
CLMN 44
GI 25 Figure(s).

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.

FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.

FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.

FIG. 4 is an illustration of the outlet channels of the device.

FIG. 5 is an illustration of a device for ***cell*** lysis.

FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.

FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.

FIG. 8 is an exploded view of a ***cell*** ***binding*** device.

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.

FIG. 10 is an illustration of types of ***obstacles***.

FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.

FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.

FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.3391. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 98%.

FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.

FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.

FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.

FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

AN 11376372 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI DEVICES AND METHODS FOR ***ENRICHMENT*** AND ALTERATION OF CELLS AND OTHER PARTICLES
IN Barber Thomas A; Carvalho Bruce L; Huang Lotien Richard; Kapur Ravi; Toner Mehmet; Vernucci Paul; Wang Zihua
PA Unassigned Or Assigned To Individual (68000)
PI US 2007026381 A1 20070201
AI US 2006-449149 20060608
RLI WO 2006-US12820 20060405 CONTINUATION PENDING
PRAI US 2005-668415P 20050405 (Provisional)
US 2005-704067P 20050729 (Provisional)
FI US 2007026381 20070201
DT Utility; Patent Application - First Publication
FS CHEMICAL APPLICATION
ED Entered STN: 2 Feb 2007
Last Updated on STN: 20 Mar 2007
CLMN 116
GI 68 Figure(s).
FIGS. 1A-1E are schematic depictions of an array that separates cells based on deterministic lateral displacement: (A) illustrates the lateral displacement of subsequent rows; (B) illustrates how fluid flowing through a gap is divide unequally around ***obstacles*** in subsequent rows; (C) illustrates how a particle with a hydrodynamic ***size*** above the critical ***size*** is displaced laterally in the device; (D) illustrates an array of cylindrical ***obstacles***; and (E) illustrates an array of elliptical ***obstacles***.
FIG. 2 is a schematic description illustrating the unequal division of the flux through a gap around ***obstacles*** in subsequent rows.
FIG. 3 is a schematic depiction of how the critical ***size*** depends on the flow profile, which is parabolic in this example.
FIG. 4 is an illustration of how shape affects the movement of particles through a device.
FIG. 5 is an illustration of how deformability affects the movement of particles through a device.
FIG. 6 is a schematic depiction of deterministic lateral displacement. Particles having a hydrodynamic ***size*** above the critical ***size*** move to the edge of the array, while particles having a hydrodynamic ***size*** below the critical ***size*** pass through the device without lateral displacement.
FIG. 7 is a schematic depiction of a three-stage device.
FIG. 8 is a schematic depiction of the maximum ***size*** and cut off ***size*** (i.e., critical ***size***) for the device of FIG. 7.
FIG. 9 is a schematic depiction of a bypass channel.
FIG. 10 is a schematic depiction of a bypass channel.
FIG. 11 is a schematic depiction of a three-stage device having a common bypass channel.
FIG. 12 is a schematic depiction of a three-stage, duplex device having a

common bypass channel.

FIG. 13 is a schematic depiction of a three stage device having a common bypass channel, where the flow through the device is substantially constant.

FIG. 14 is a schematic depiction of a three stage, duplex device having a common bypass channel, where the flow through the device is substantially constant.

FIG. 15 is a schematic depiction of a three stage device having a common bypass channel, where the fluidic resistance in the bypass channel and the adjacent stage are substantially constant.

FIG. 16 is a schematic depiction of a three stage, duplex device having a common bypass channel, where the fluidic resistance in the bypass channel and the adjacent stage are substantially constant.

FIG. 17 is a schematic depiction of a three-stage device having two, separate bypass channels.

FIG. 18 is a schematic depiction of a three stage device having two, separate bypass channels, which are in arbitrary configuration.

FIG. 19 is a schematic depiction of a three-stage, duplex device having three, separate bypass channels.

FIG. 20 is a schematic depiction of a three stage device having two, separate bypass channels, wherein the flow through each stage is substantially constant.

FIG. 21 is a schematic depiction of a three stage, duplex device having three, separate bypass channels, wherein the flow through each stage is substantially constant.

FIG. 22 is a schematic depiction of a flow extracting boundary.

FIG. 23 is a schematic depiction of a flow feeding boundary.

FIG. 24 is a schematic depiction of a flow-feeding boundary, including a bypass channel.

FIG. 25 is a schematic depiction of two flow-feeding boundaries flanking a central bypass channel.

FIG. 26 is a schematic depiction of a device having four channels that act as on-chip flow resistors.

FIGS. 27 and 28 are schematic depictions of the effect of on-chip resistors on the relative width of two fluids flowing in a device.

FIG. 29 is a schematic depiction of a duplex device having a common inlet for the two outer regions.

FIG. 30A is a schematic depiction of a multiple arrays on a device. FIG. 30B is a schematic depiction of multiple arrays with common inlets and product outlets on a device.

FIG. 31 is a schematic depiction of a multi-stage device with a small footprint.

FIG. 32 is a schematic depiction of blood passing through a device.

FIG. 33 is a graph illustrating the hydrodynamic ***size*** distribution of blood cells.

FIGS. 34A-34D are schematic depictions of moving a particle from a sample to a buffer in a single stage (A), three-stage (B), duplex (C), or three stage duplex (D) device.

FIG. 35A is a schematic depiction of a two stage device employed to move a particle from blood to a buffer to produce three products. FIG. 35B is a schematic graph of the maximum ***size*** and cut off ***size*** of the two stages.

FIG. 35C is a schematic graph of the composition of the three products.

FIG. 36 is a schematic depiction of a two-stage device for alteration, where each stage has a bypass channel.

FIG. 37 is a schematic depiction of the use of fluidic channels to connect two stages in a device.

FIG. 38 is a schematic depiction of the use of fluidic channels to connect two stages in a device, wherein the two stages are configured as a small footprint array.

FIG. 39A is a schematic depiction of a two stage device having a bypass channel that accepts output from both stages. FIG. 39B is a schematic graph of the range of product sizes achievable with this device.

FIG. 40 is a schematic depiction of a two stage device for alteration having bypass channels that flank each stage and empty into the same outlet.

FIG. 41 is a schematic depiction of a device for the sequential movement and alteration of particles.

FIG. 42A is a photograph of a device of the invention. FIGS. 42B-42E are depictions the mask used to fabricate a device of the invention. FIG. 42F is a series of photographs of the device containing blood and buffer.

FIGS. 43A-43F are typical histograms generated by the hematology analyzer

from a blood sample and the waste (buffer, plasma, ***red*** ***blood*** ***cells***, and platelets) and product (buffer and nucleated cells) fractions generated by the device of FIG. 42.
FIGS. 44A-44D are depictions the mask used to fabricate a device of the invention.
FIGS. 45A-45D are depictions the mask used to fabricate a device of the invention.
FIG. 46A is a micrograph of a sample ***enriched*** in fetal ***red*** ***blood*** ***cells*** . FIG. 46B is a micrograph of maternal red blood ***cell*** waste.
FIG. 47 is a series of micrographs showing the positive identification of male fetal cells (Blue-nucleus, Red-X chromosome, Green-Y chromosome).
FIG. 48 is a series of micrographs showing the positive identification of sex and trisomy 21.
FIGS. 49A-49D are depictions the mask used to fabricate a device of the invention.
FIGS. 50A-50C are electron micrographs of the device of FIG. 49.
FIGS. 51A-51D are depictions the mask used to fabricate a device of the invention.
FIGS. 52A-52F are electron micrographs of the device of FIG. 51.
FIGS. 53A-53F are electron micrographs of the device of FIG. 45.
FIGS. 54A-54D are depictions the mask used to fabricate a device of the invention.
FIGS. 55A-55S are electron micrographs of the device of FIG. 54.
FIGS. 56A-56C are electron micrographs of the device of FIG. 44.
FIG. 57 is a flowchart describing the isolation of fetal red blood ***cell*** nuclei.
FIG. 58 is a schematic graph of the course of lysis of cells in a maternal blood sample.
FIG. 59 is a schematic diagram of a ***microfluidic*** method to ***enrich*** the cells of interest and preferentially lyse the cells of interest in the ***enriched*** sample. The sample is first ***enriched*** by ***size*** based direction of cells of interest into a preferred channel, and the cells of interest are then selectively lysed by controlling their residence time in a lysis solution.
FIG. 60 is a schematic diagram of a ***microfluidic*** method of sizebased isolation of the nuclei of the lysed cells of interest from non-lysed whole cells of non-interest. The cells of noninterest are directed into the waste, while the nuclei are retained in the desired product stream.
FIG. 61 is a flowchart describing an alternate method for the separation of fetal nuclei from maternal white blood cells.
FIG. 62 is a schematic diagram of a device of the invention employing a substantially constant gap width and flow-feeding and flow-extracting boundaries.
FIG. 63a is a schematic depiction of a manifold of the invention. FIG. 63b is a photograph of a manifold of the invention.
FIG. 64 is a graph of the percentage of viable cells as a function of exposure to a hypotonic lysis solution.
FIG. 65 is a graph of hemolysis of whole blood as a function of time in a lysis buffer.
FIG. 66a is a table that illustrates the nuclei recovery after Cytospin using Carnoy's fix solution total ***cell*** lysis procedure as described herein.
FIG. 66b is a series of fluorescent micrographs showing an example of nuclei FISH results using Carnoy's fix mediated total ***cell*** lysis. The nuclei are FISHed for X (aqua), Y (green) and Y (red) and counterstained with DAPI.
FIG. 67 is a flowchart detailing various options for lysis of cells and nuclei.

L10 ANSWER 7 OF 19 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11185565 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA Unassigned Or Assigned To Individual (68000)
PPA General Hospital Corp The (Probable)
PI US 2006134599 A1 20060622
AI US 2003-529453 20030929
WO 2003-US30965 20030929
20051219 PCT 371 date

20051219 PCT 102(e) date

PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)

FI US 2006134599 20060622

DT Utility; Patent Application - First Publication

FS CHEMICAL APPLICATION

ED Entered STN: 24 Jun 2006 Last Updated on STN: 24 Jun 2006

CLMN 69

GI 32 Figure(s).

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.

FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.

FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.

FIG. 4 is an illustration of the outlet channels of the device.

FIG. 5 is an illustration of a device for ***cell*** lysis.

FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.

FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.

FIG. 8 is an exploded view of a ***cell*** ***binding*** device.

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.

FIG. 10 is an illustration of types of ***obstacles***.

FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%.

FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array.

FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.

FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure drop.

FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.

FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.3391. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 99%.

FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 mL/hr.

FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.

FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.

FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series.
FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention.
FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation.
FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

L10 ANSWER 8 OF 10 USPATFULL on STN
AN 2008:130394 USPATFULL <<LOGINID::20080526>>
TI SELECTION OF CELLS USING BIOMARKERS
IN Kapur, Ravi, Stoughton, MA, UNITED STATES
Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
Wang, Zihua, Newton, MA, UNITED STATES
PI US 20081113358 A1 20080515
AI US 2007-830546 A1 20070730 (11)
PRAI US 2006-820778P 20060728 (60)
DT Utility
FS APPLICATION
LN.CNT 3370
INCL INCLM: 435.6
NCL NCLM: 435.6
IC IPCI C12Q0001-68 [I,A]

L10 ANSWER 9 OF 10 USPATFULL on STN
AN 2008:76951 USPATFULL <<LOGINID::20080526>>
TI DC dielectrophoresis ***microfluidic*** apparatus, and applications of same
IN Li, Dongqing, Antioch, TN, UNITED STATES
PA Vanderbilt University, Nashville, TN, UNITED STATES (U.S. corporation)
PI US 2008067068 A1 20080320
AI US 2006-523782 A1 20060919 (11)
DT Utility
FS APPLICATION
LN.CNT 2209
INCL INCLM: 204/451.000
INCLS: 204/601.000
NCL NCLM: 204/451.000
NCLS: 204/601.000
IC IPCI C07K0001-26 [I,A]; C07K0001-00 [I,C*]; G01N0027-00 [I,A]
IPCR C07K0001-00 [I,C]; C07K0001-26 [I,A]; G01N0027-00 [I,C]; G01N0027-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10 ANSWER 10 OF 10 USPATFULL on STN
AN 2007:68524 USPATFULL <<LOGINID::20080526>>
TI System for size based separation and analysis
IN Kapur, Ravi, Stoughton, MA, UNITED STATES
Toner, Mehmet, Wellesley, MT, MA, UNITED STATES
Huang, Lotien R., Brookline, MA, UNITED STATES
Barber, Tom, Cambridge, MA, UNITED STATES
Carvalho, Bruce, Watertown, MA, UNITED STATES
Gray, Darren, Brookline, MA, UNITED STATES

```
PI      US 2007059781          A1   20070315
AI      US 2005-229336         A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2262
INCL    INCLM: 435/007.210
        INCLS: 435/287.200; 702/019.000
NCL     NCLM:  435/007.210
        NCLS:  435/287.200; 702/019.000
IC      IPCI   G01N0033-567 [I,A]; G06F0019-00 [I,A]; C12M0003-00 [I,A]
        IPCR   G01N0033-567 [I,C]; G01N0033-567 [I,A]; C12M0003-00 [I,C];
               C12M0003-00 [I,A]; G06F0019-00 [I,C]; G06F0019-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
L10  ANSWER 11 OF 19  USPATFULL on STN
AN      2007:68527  USPATFULL <<LOGINID::20080526>>
TI      Kits for Prenatal Testing
IN      Grigham, Michael, Richmond, VA, UNITED STATES
        Kapur, Ravi, Stoughton, MA, UNITED STATES
        Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
PI      US 2007059774          A1   20070315
AI      US 2005-229037         A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2316
INCL    INCLM: 435/007.200
NCL     NCLM:  435/007.200
IC      IPCI   G01N0033-567 [I,A]; G01N0033-53 [I,A]
        IPCR   G01N0033-567 [I,C]; G01N0033-567 [I,A]; G01N0033-53 [I,C];
               G01N0033-53 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
L10  ANSWER 12 OF 19  USPATFULL on STN
AN      2007:68473  USPATFULL <<LOGINID::20080526>>
TI      Business methods for prenatal Diagnosis
IN      Grigham, Michael, Richmond, VA, UNITED STATES
        Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
        Tompkins, Ron, Boston, MA, UNITED STATES
        Schmidt, Martin, Reading, MA, UNITED STATES
        Kapur, Ravi, Stoughton, MA, UNITED STATES
PI      US 2007059719          A1   20070315
AI      US 2005-229332         A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2256
INCL    INCLM: 435/006.000
        INCLS: 705/002.000
NCL     NCLM:  435/006.000
        NCLS:  705/002.000
IC      IPCI   C12Q0001-68 [I,A]; G06Q0050-00 [I,A]
        IPCR   C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; G06Q0050-00 [I,C];
               G06Q0050-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
L10  ANSWER 13 OF 19  USPATFULL on STN
AN      2007:68472  USPATFULL <<LOGINID::20080526>>
TI      Systems and methods for ***enrichment*** of analytes
IN      Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
        Kapur, Ravi, Stoughton, MA, UNITED STATES
PI      US 2007059718          A1   20070315
AI      US 2005-229328         A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2296
INCL    INCLM: 435/006.000
        INCLS: 435/069.100
NCL     NCLM:  435/006.000
        NCLS:  435/069.100
IC      IPCI   C12Q0001-68 [I,A]; C12P0021-06 [I,A]
        IPCR   C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; C12P0021-06 [I,C];
               C12P0021-06 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.
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L10 ANSWER 14 OF 19 USPATFULL on STN
AN 2007:68470 USPATFULL <<LOGINID::20080526>>
TI Methods for detecting fetal abnormality
IN Balis, Ulysses, Peabody, MA, UNITED STATES
Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
Kapur, Ravi, Stoughton, MA, UNITED STATES
Walsh, John, Auburndale, MA, UNITED STATES
PI US 2007059716 A1 20070315
AI US 2005-228454 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2200
INCL INCLM: 435/006.000
INCLS: 702/020.000
NCL NCLM: 435/006.000
NCLS: 702/020.000
IC IPCI C12Q0001-68 [I,A]; G06F0019-00 [I,A]
IPCR C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; G06F0019-00 [I,C];
G06F0019-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10 ANSWER 15 OF 19 USPATFULL on STN
AN 2007:68437 USPATFULL <<LOGINID::20080526>>
TI Veterinary diagnostic system
IN Barber, Tom, Cambridge, MA, UNITED STATES
Huang, Lotien R., Brookline, MA, UNITED STATES
Gray, Darren, Brookline, MA, UNITED STATES
Kapur, Ravi, Stoughton, MA, UNITED STATES
PI US 2007059683 A1 20070315
AI US 2005-229359 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2331
INCL INCLM: 435/005.000
INCLS: 435/006.000; 435/007.200; 977/902.000; 977/924.000
NCL NCLM: 435/005.000
NCLS: 435/006.000; 435/007.200; 977/902.000; 977/924.000
IC IPCI C12Q0001-70 [I,A]; C12Q0001-68 [I,A]; G01N0033-567 [I,A];
G01N0033-53 [I,A]
IPCR C12Q0001-70 [I,C]; C12Q0001-70 [I,A]; C12Q0001-68 [I,C];
C12Q0001-68 [I,A]; G01N0033-53 [I,C]; G01N0033-53 [I,A];
G01N0033-567 [I,C]; G01N0033-567 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10 ANSWER 16 OF 19 USPATFULL on STN
AN 2007:68434 USPATFULL <<LOGINID::20080526>>
TI System for cell ***enrichment***
IN Kapur, Ravi, Stoughton, MA, UNITED STATES
Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
Huang, Lotien R., Brookline, MA, UNITED STATES
PI US 2007059680 A1 20070315
AI US 2005-228462 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2356
INCL INCLM: 435/004.000
INCLS: 435/005.000; 435/287.100; 435/006.000
NCL NCLM: 435/004.000
NCLS: 435/005.000; 435/006.000; 435/287.100
IC IPCI C12Q0001-00 [I,A]; C12Q0001-70 [I,A]; C12Q0001-68 [I,A];
C12M0003-00 [I,A]
IPCR C12Q0001-00 [I,C]; C12Q0001-00 [I,A]; C12M0003-00 [I,C];
C12M0003-00 [I,A]; C12Q0001-68 [I,C]; C12Q0001-68 [I,A];
C12Q0001-70 [I,C]; C12Q0001-70 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10 ANSWER 17 OF 19 USPATFULL on STN
AN 2004:282265 USPATFULL <<LOGINID::20080526>>
TI ***Microfluidic*** particle-analysis systems
IN Daridon, Antoine, Belmont, CA, UNITED STATES
PA Fluidigm Corporation, South San Francisco, CA, 94080 (U.S. corporation)

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PI     US 2004229249       A1  20041118
AI     US 2003-640510      A1  20030813 (10)
RLI    Continuation-in-part of Ser. No. US 2003-405092, filed on 31 Mar 2003,
       ABANDONED
PRAI   US 2002-369538P     20020401 (60)
       US 2002-378464P     20020506 (60)
DT     Utility
FS     APPLICATION
LN.CNT 6336
INCL   INCLM: 435/305.200
NCL    NCLM:  435/305.200
IC     [7]
       ICM    C12M001-32
       IPCI   C12M0001-32 [ICM,7]
       IPCR   G01N0037-00 [I,C*]; G01N0037-00 [I,A]; B01L0003-00 [I,C*];
              B01L0003-00 [I,A]; C12M0001-00 [I,C*]; C12M0001-00 [I,A];
              C12M0001-34 [I,C*]; C12M0001-34 [I,A]; C12M0003-00 [I,C*];
              C12M0003-00 [I,A]; C12N0001-00 [I,C*]; C12N0001-00 [I,A];
              C12Q0001-02 [I,C*]; C12Q0001-02 [I,A]; G01N0015-02 [N,C*];
              G01N0015-02 [N,A]; G01N0015-14 [I,C*]; G01N0015-14 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10  ANSWER 18 OF 19  USPATFULL on STN
AN     2004:286218  USPATFULL <<LOGINID::20080526>>
TI       ***Microfluidic***  particle-analysis systems
IN     Chou, Hou-Pu, Sunnyvale, CA, UNITED STATES
       Daridon, Antoine, Belmont, CA, UNITED STATES
       Farrell, Kevin, San Francisco, CA, UNITED STATES
       Fowler, Brian, Foster City, CA, UNITED STATES
       Liau, Yish-Hann, San Jose, CA, UNITED STATES
       Manger, Ian D., San Francisco, CA, UNITED STATES
       Nassef, Hany Ramez, San Mateo, CA, UNITED STATES
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PI     US 2004224380       A1  20041111
AI     US 2003-724963      A1  20031211 (10)
RLI    Continuation of Ser. No. US 2003-405953, filed on 1 Apr 2003, PENDING
PRAI   US 2002-369538P     20020401 (60)
       US 2002-378464P     20020506 (60)
DT     Utility
FS     APPLICATION
LN.CNT 6605
INCL   INCLM: 435/029.000
       INCLS: 435/288.500; 422/068.100; 436/063.000
NCL    NCLM:  435/029.000
       NCLS:  422/068.100; 435/288.500; 436/063.000
IC     [7]
       ICM    G01N033-00
       ICS    C12M001-34; C12Q001-02
       IPCI   G01N0033-00 [ICM,7]; C12M0001-34 [ICS,7]; C12Q0001-02 [ICS,7]
       IPCR   B01L0003-00 [I,C*]; B01L0003-00 [I,A]; G01N0015-02 [N,C*];
              G01N0015-02 [N,A]; G01N0015-14 [I,C*]; G01N0015-14 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L10  ANSWER 19 OF 19  USPATFULL on STN
AN     2004:94825  USPATFULL <<LOGINID::20080526>>
TI       ***Microfluidic***  particle-analysis systems
IN     Chou, Hou-Pu, Sunnyvale, CA, UNITED STATES
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PI     US 2004072278       A1  20040415
       US 7312085          B2  20071225
AI     US 2003-405953      A1  20030401 (10)
PRAI   US 2002-369538P     20020401 (60)
       US 2002-378464P     20020506 (60)
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DT Utility
FS APPLICATION
LN.CNT 6606
INCL INCLM: 435/029.000
INCLS: 422/068.100; 436/063.000; 435/288.500
NCL NCLM: 436/043.000; 435/029.000
NCLS: 422/050.000; 422/068.100; 422/081.000; 422/082.000; 422/100.000; 422/101.000; 422/102.000; 422/103.000; 422/104.000; 435/004.000; 435/325.000; 435/282.000; 435/283.000; 435/284.000; 435/404.000; 435/405.000; 436/063.000; 436/180.000; 435/288.500
IC [7]
ICM G01N033-00
ICS C12M001-34; C12Q001-02
IPCI G01N0033-00 [ICM,7]; C12M0001-34 [ICS,7]; C12Q0001-02 [ICS,7]
IPCI-2 G01N0035-00 [I,A]; G01N0033-48 [I,A]; G01N0001-10 [I,A]; G01N0015-06 [I,A]; G01N0033-00 [I,A]
IPCR G01N0035-00 [I,C]; G01N0035-00 [I,A]; B01L0003-00 [I,C*]; B01L0003-00 [I,A]; G01N0001-10 [I,C]; G01N0001-10 [I,A]; G01N0015-02 [N,C*]; G01N0015-02 [N,A]; G01N0015-06 [I,C]; G01N0015-06 [I,A]; G01N0015-14 [I,C*]; G01N0015-14 [I,A]; G01N0033-00 [I,C]; G01N0033-00 [I,A]; G01N0033-48 [I,C]; G01N0033-48 [I,A]

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

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L19 NOT FOUND
The L-number entered has not been defined in this session, or it has been deleted. To see the L-numbers currently defined in this session, enter DISPLAY HISTORY at an arrow prompt (=>).

=> d l10 19 ab

L10 ANSWER 19 OF 19 USPATFULL on STN
AB The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or detection of particles, such as cells and/or beads. The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or analysis of particles, such as cells, viruses, organelles, beads, and/or vesicles. The invention also provides ***microfluidic*** mechanisms for carrying out these manipulations and analyses. These mechanisms may enable controlled input, movement/positioning, retention/localization, treatment, measurement, release, and/or output of particles. Furthermore, these mechanisms may be combined in any suitable order and/or employed for any suitable number of times within a system. Accordingly, these combinations may allow particles to be sorted, cultured, mixed, treated, and/or assayed, among others, as single particles, mixed groups of particles, arrays of particles, heterogeneous particle sets, and/or homogeneous particle sets, among others, in series and/or in parallel. In addition, these combinations may enable ***microfluidic*** systems to be reused. Furthermore, these combinations may allow the response of particles to treatment to be measured on a shorter time scale than was previously possible. Therefore, systems of the invention may allow a broad range of ***cell*** and particle assays, such as drug screens, ***cell*** characterizations, research studies, and/or clinical analyses, among others, to be scaled down to ***microfluidic*** ***size***. Such scaled-down assays may use less sample and reagent, may be less labor intensive, and/or may be more informative than comparable macrofluidic assays.

=> d l10 19 kwic

L10 ANSWER 19 OF 19 USPATFULL on STN
TI ***Microfluidic*** particle-analysis systems
AB The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or detection of particles, such as cells and/or beads. The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or analysis of particles, such as cells, viruses, organelles, beads, and/or vesicles. The invention also provides

microfluidic mechanisms for carrying out these manipulations and analyses. These mechanisms may enable controlled input, movement/positioning, retention/localization, treatment, measurement, release, and/or ... heterogeneous particle sets, and/or homogeneous particle sets, among others, in series and/or in parallel. In addition, these combinations may enable ***microfluidic*** systems to be reused. Furthermore, these combinations may allow the response of particles to treatment to be measured on a shorter time scale than was previously possible. Therefore, systems of the invention may allow a broad range of ***cell*** and particle assays, such as drug screens, ***cell*** characterizations, research studies, and/or clinical analyses, among others, to be scaled down to ***microfluidic*** ***size***. Such scaled-down assays may use less sample and reagent, may be less labor intensive, and/or may be more informative than ...,

PARN ... and therein. This application further claims priority under 35 U.S.C. sctn. 120 as a continuation-in-part of the non-provisional patent application titled " ***Microfluidic*** Particle-Analysis Systems" by Chou et al., filed on Mar. 31, 2003 (Atty. Docket No.: 139F.310US), which is hereby incorporated by ...

SUMM [0004] The invention relates to systems for the manipulation and/or detection of particles. More particularly, the invention relates to ***microfluidic*** systems for the manipulation and/or detection of particles, such as cells and/or beads.

SUMM [0007] The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or detection of particles, such as cells and/or beads.

DRWD ... 1 is a flow chart showing potential temporal relationships between method steps for manipulation and/or detection of particles in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0009] FIG. 2A is a top plan view of a ***microfluidic*** system for retaining and analyzing a subset of input particles, in accordance with aspects of the invention.

DRWD [0010] FIG. 2B is a top plan view of another ***microfluidic*** system for retaining and analyzing a subset of input particles, in accordance with aspects of the invention.

DRWD [0011] FIG. 3 is a fragmentary, top plan view of yet another ***microfluidic*** system for retaining and analyzing a subset of input particles, in accordance with aspects of the invention.

DRWD [0012] FIG. 5 is a fragmentary, top plan view of a ***microfluidic*** system for positioning and retaining a group of particles, and for perfusing the retained group with selected reagents, in accordance. .

DRWD [0020] FIG. 11A is a fragmentary, top plan view of a ***microfluidic*** system for measuring cell-cell communication, based on a duplicated version of the system of FIG. 8, in accordance with aspects ...

DRWD ... 11C is a top plan view of a two-dimensional array of particle capture chambers that may be used in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0023] FIG. 12 is a fragmentary, top plan view of a ***microfluidic*** system for retaining and perfusing two sets of particles in parallel, in accordance with aspects of the invention.

DRWD [0025] FIG. 13A is a top plan view of a ***microfluidic*** system for retaining two particles at spaced sites in a channel and perfusing the retained particles with distinct reagents, in ...

DRWD [0026] FIG. 13I is a fragmentary, top plan view of a ... ***microfluidic*** system having separately addressable sets of linear trap arrays, in accordance with aspects of the invention.

DRWD [0027] FIG. 14 is a top plan view of a ***microfluidic*** system for retaining an array of particles in series and for perfusing members of this array separately and in parallel.

DRWD [0040] FIG. 17 is a fragmentary, top plan view of a ***microfluidic*** ***device*** for forming an array of single particles or groups of particles, in accordance with aspects of the invention.

DRWD [0041] FIG. 18 is a pair of fragmentary, top plan schematic views of a ***microfluidic*** ***device*** for forming an array of retained particles that may be transferred to an array of separate sites, illustrating particle retention.

DRWD [0042] FIG. 19 is a pair of fragmentary, top plan schematic views of another ***microfluidic*** ***device*** for forming an array of retained particles that may be transferred to an array of separate

sites, illustrating particle retention

DRWD [0043] FIG. 20 is fragmentary, top plan schematic view of yet another ***microfluidic*** ***device*** for forming an array of retained particles that may be transferred to an array of separate sites, in accordance with

DRWD [0044] FIG. 21 is a composite of top plan and sectional views showing selected portions of a ***microfluidic*** system for retaining particles using a particle retention chamber that is fully spaced from the floor of the system, in accordance

DRWD FIG. 22 is a composite of top plan and sectional views, and a photographic image, showing selected portions of a ***microfluidic*** system for retaining particles using a particle retention chamber that is partially spaced from the floor of the system, in accordance

DRWD FIG. 23 is a composite of top plan and sectional views, and two photographic images, showing selected portions of another ***microfluidic*** system for retaining particles using a particle retention chamber that is fully spaced from the floor of the system, in accordance

DRWD [0047] FIG. 24 is a fragmentary, top plan view of a reusable ***microfluidic*** system for repeated retention, treatment, and release of single particles, in accordance with aspects of the invention.

DRWD [0049] FIG. 26 is a fragmentary, top plan view of a reusable ***microfluidic*** system for repeated retention, treatment, and release of groups of particles, in accordance with aspects of the invention.

DRWD [0054] FIG. 31 is a top plan view of a ***microfluidic*** system having a sorting mechanism based on centrifugal force, in accordance with aspects of the invention.

DRWD [0056] FIG. 33 is a fragmentary, top plan view of another ***microfluidic*** system having a sorting mechanism based on centrifugal force, in accordance with aspects of the invention.

DRWD [0057] FIG. 34 is a top plan view of a yet another ***microfluidic*** system having a sorting mechanism based on centrifugal force, in accordance with aspects of the invention.

DRWD [0062] FIGS. 39-43 are top plan composite views of various cell chamber networks for ***microfluidic*** manipulation of cells, in accordance with aspects of the invention.

DRWD [0063] FIG. 44 is a top plan view of a ***microfluidic*** system with a parallel array of separate, isolatable cell-chamber networks, in accordance with aspects of the invention.

DRWD [0064] FIG. 45 is a top plan view of a ***microfluidic*** system with an isolatable cell chamber that may be fed or bypassed by a parallel fluidic circuit, in accordance with

DRWD [0065] FIG. 46 is a top plan view of a ***microfluidic*** system having a cell chamber that forms a loop, in accordance with aspects of the invention.

DRWD [0066] FIG. 47 is a top plan view of a ***microfluidic*** system in which particle and reagent networks intersect at a common cell chamber, in accordance with aspects of the invention.

DRWD [0071] FIG. 50C is a top plan view of a ***cell*** chamber having two distinct compartments connected by a set of radially arrayed ***size*** -selective channels, in accordance with aspects of the invention.

DRWD [0073] FIG. 51 is an isometric schematic view of a ***microfluidic*** system for performing electrophysiological analysis on an array of cells, in accordance with aspects of the invention.

DRWD [0074] FIG. 52 is a top plan view of a ***microfluidic*** system for performing electrophysiological analysis on a single cell, in accordance with aspects of the invention.

DRWD [0075] FIG. 53 is a fragmentary top plan view of a ***microfluidic*** system related to the system of FIG. 52, showing a modified focusing mechanism, in accordance with aspects of the invention.

DRWD [0081] FIG. 59 is an abstracted view of a ***microfluidic*** ***device*** for performing patch clamp analysis of cells, in accordance with aspects of the invention.

DRWD [0082] FIG. 60 is a fragmentary top plan view of a ***microfluidic*** ***device*** for performing patch clamp analysis of multiple individual cells, in accordance with aspects of the invention.

DRWD [0084] FIG. 62 is a fragmentary side elevation view of a ***microfluidic*** mold spin coated with a first layer of patternable, selectively removable material, in accordance with aspects of the

invention.

DRWD and measured signal plus noise without (top) and with (bottom) implementation of the modulation demodulation method of FIG. 71B in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD intensity versus time prior to and during cycles of exposure of a biotinylated bead to a streptavidin dye conjugate in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD ionomycin to a retained cell that was preloaded with a calcium sensor dye, using the method of FIG. 71B in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0099] FIG. 71E is a graph of measured fluorescence intensity versus time at a position in a ***microfluidic*** system prior to and during exposure to a dye, in accordance with aspects of the invention.

DRWD [0100] FIG. 72 is a time lapse set of photographic images recording size selective flow of blood cells through a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0101] FIG. 73 is diagram showing the structure of biotin and its mode of ***binding*** to streptavidin.

DRWD [0102] FIG. 74 is a time lapse set of photographic images recording interaction of specific ***binding*** pairs on beads in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0103] FIG. 75 is a time lapse set of photographic images recording stimulation of ion flux in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0104] FIG. 76 is a time lapse set of photographic images recording apoptosis and necrosis in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0106] FIG. 79 is a photographic image recording successful staining of a cell's membrane in a non-***microfluidic*** environment.

DRWD 80 is a time lapse set of photographic images recording retention of a single cell at a preselected site in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD is a time lapse set of photographic images recording retention of a group of cells at a preselected site in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0110] FIG. 83 is a time lapse set of photographic images recording fixation and staining of a retained cell in a ***microfluidic*** system, in accordance with aspects of the invention.

DRWD [0111] FIG. 84 is a top plan view of a ***microfluidic*** system for analyzing a size-selected set of cells, in which the system includes serially disposed filtration and retention mechanisms, a

DRWD [0112] FIG. 85 is another top plan view of the ***microfluidic*** system of FIG. 84, showing identifying labels for reservoirs and valves, in accordance with aspects of the invention.

DETD [0118] The invention provides systems, including apparatus, methods, and kits, for the ***microfluidic*** manipulation and/or analysis of particles, such as cells, viruses, organelles, beads, and/or vesicles. The invention also provides ***microfluidic*** mechanisms for carrying out these manipulations and analyses. These mechanisms may enable controlled input, movement/positioning, retention/localization, treatment, measurement, release, and/or . . . heterogeneous particle sets, and/or homogeneous particle sets, among others, in series and/or in parallel. In addition, these combinations may enable ***microfluidic*** systems to be reused. Furthermore, these combinations may allow the response of particles to treatment to be measured on a shorter time scale than was previously possible. Therefore, systems of the invention may allow a broad range of ***cell*** and particle assays, such as drug screens, ***cell*** characterizations, research studies, and/or clinical analyses, among others, to be scaled down to ***microfluidic*** ***size***. Such scaled down assays may use less sample and reagent, may be less labor intensive, and/or may be more informative than . . .

DETD [0120] Further aspects of the invention are described in the following sections: (I) ***microfluidic*** systems, (II) physical structures of fluid networks, (III) particles, (IV) input mechanisms, (V) positioning mechanisms, (VI) retention mechanisms, (VII) treatment. . .

DETD [0121] ***Microfluidic*** Systems
DETD [0122] Particle manipulations and analyses are performed in ***microfluidic*** systems. A ***microfluidic*** system generally comprises any system in which very small volumes of fluid are stored and manipulated, generally less than about 500 mu L, typically less than about 100 mu L, and more typically less than about 10 mu L. ***Microfluidic*** systems carry fluid in predefined paths through one or more ***microfluidic*** passages. A ***microfluidic*** passage may have a minimum dimension, generally height or width, of less than about 200, 100, or 50 mu m. Passages

DETD [0124] ***Microfluidic*** systems may include one or more sets of passages that interconnect to form a generally closed ***microfluidic*** network. Such a ***microfluidic*** network may include one, two, or more openings at network termini, or intermediate to the network, that interface with the external world. Such openings may receive, store, and/or dispense fluid. Dispensing fluid may be directly into the ***microfluidic*** network or to sites external the ***microfluidic*** system. Such openings generally function in input and/or output mechanisms, described in more detail in Sections IV and X below.

DETD [0125] ***Microfluidic*** systems also may include any other suitable features or mechanisms that contribute to fluid, reagent, and/or particle manipulation or analysis. For example, ***microfluidic*** systems may include regulatory or control mechanisms that determine aspects of fluid flow rate and/or path. Valves and/or pumps that may participate in such regulatory mechanisms are described in more detail below in Section II. Alternatively, or in addition, ***microfluidic*** systems may include mechanisms that determine, regulate, and/or sense fluid temperature, fluid pressure, fluid flow rate, exposure to light, exposure to electric fields, magnetic field strength, and/or the like. Accordingly, ***microfluidic*** systems may include heaters, coolers, electrodes, lenses, gratings, light sources, pressure sensors, pressure transducers, microprocessors, microelectronics, and/or so on. Furthermore, each ***microfluidic*** system may include one or more features that act as a code to identify a given system. The features may

DETD [0127] ***Microfluidic*** systems may be formed of any suitable material or combination of suitable materials. Suitable materials may include elastomers, such as

DETD [0128] Exemplary materials for ***microfluidic*** systems are described in more detail in the patent applications listed above under Cross-References, which are incorporated herein by reference.

DETD [0130] ***Microfluidic*** systems, also referred to as chips, may have any suitable structure. Such systems may be fabricated as a unitary structure

DETD substantially planar layer that functions as a substrate layer, in some cases contributing a floor portion to some or all ***microfluidic*** passages.

DETD [0132] Components of a ***microfluidic*** system may be fabricated by any suitable mechanism, based on the desired application for the system and on materials used. material by micromachining, etching, soft lithography, material deposition, cutting, and/or punching, among others. Alternatively, or in addition, components of a ***microfluidic*** system may be fabricated without a mold by etching, micromachining, cutting, punching, and/or material deposition.

DETD [0133] ***Microfluidic*** components may be fabricated separately, joined, and further modified as appropriate. For example, when fabricated as distinct layers, ***microfluidic*** components may be bonded, generally face to face. These separate components may be surface treated, for example, with reactive chemicals to modify surface chemistry, with particle ***binding*** agents, with reagents to facilitate analysis, and/or so on. Such surface treatment may be localized to discrete portions of the surface.

DETD [0134] Exemplary methods for fabricating ***microfluidic*** systems are described in more detail in the patent applications identified above under Cross-References, which are incorporated herein by reference.

DETD [0137] ***Microfluidic*** systems may include any suitable structure(s) for the integrated manipulation of small volumes of fluid, including moving and/or storing fluid

DETD suitable path, channel, or duct through, over, or along which materials (e.g., fluid, particles, and/or reagents) may pass in a ***microfluidic*** system. Collectively, a set of fluidically

communicating passages, generally in the form of channels, may be referred to as a ***microfluidic*** network. In some cases, passages may be described as having surfaces that form a floor, a roof, and walls. Passages

DETD [0141] Passages may branch, join, and/or dead-end to form any suitable ***microfluidic*** network. Accordingly, passages may function in particle positioning, sorting, retention, treatment, detection, propagation, storage, mixing, and/or release, among others.

DETD and/or output reservoirs. Input reservoirs may store materials (e.g., fluid, particles, and/or reagents) prior to inputting the materials to a ***microfluidic*** network(s) portion of a chip. By contrast, intermediate reservoirs may store materials during and/or between processing operations. Finally, output reservoirs

DETD [0151] ***Microfluidic*** systems may be used to manipulate and/or analyze particles. A particle generally comprises any object that is small enough to be inputted and manipulated within a ***microfluidic*** network in association with fluid, but that is large enough to be distinguishable from the fluid. Particles, as used here

DETD [0155] Cells used as particles in ***microfluidic*** systems may have any suitable origin, genetic background, state of health, state of fixation, membrane permeability, pretreatment, and/or population purity, . . . have intact membranes, and/or permeabilized/disrupted membranes to allow uptake of ions, labels, dyes, ligands, etc., or to allow release of ***cell*** contents. Cells may have been pretreated before introduction into a ***microfluidic*** system by any suitable processing steps. Such processing steps may include modulator treatment, transfection (including infection, injection, particle bombardment, lipofection, . . . or labels, and/or so on. Furthermore, cells may be a monoculture, generally derived as a clonal population from a single ***cell*** or a small set of very similar cells; may be presorted by any suitable mechanism such as affinity, ***binding***, FACS, drug selection, etc.; and/or may be a mixed or heterogeneous population of distinct ***cell*** types.

DETD [0167] Viruses may be manipulated and/or analyzed as particles in ***microfluidic*** systems. Viruses generally comprise any microscopic/submicroscopic parasites of cells (animals, plants, fungi, protists, and/or bacteria) that include a protein and/or. . . .

DETD [0169] Organelles may be manipulated and/or analyzed in ***microfluidic*** systems. Organelles generally comprise any particulate component of a cell. For example, organelles may include nuclei, Golgi apparatus, lysosomes, endosomes,

DETD mixtures, phages, viruses, and/or cells, among others. For example, the beads may be associated with a member of a specific ***binding*** pair (see Section VI), such as a receptor, a ligand, a nucleic acid, a member of a compound library, and/or

DETD [0177] ***Microfluidic*** systems may include one or more input mechanisms that interface with the ***microfluidic*** network(s). An input mechanism generally comprises any suitable mechanism for inputting material(s) (e.g., particles, fluid, and/or reagents) to a ***microfluidic*** network of a ***microfluidic*** chip, including selective (that is, component-by-component) and/or bulk mechanisms.

DETD [0178] The input mechanism may receive material from internal sources, that is, reservoirs that are included in a ***microfluidic*** chip, and/or external sources, that is, reservoirs that are separate from, or external to, the chip.

DETD [0186] ***Microfluidic*** systems may include one or more positioning mechanisms. A positioning mechanism generally comprises any mechanism for placing particles at preselected

DETD particle position longitudinally and/or transversely. The term "longitudinal position" denotes position parallel to or along the long axis of a ***microfluidic*** channel and/or a fluid flow stream within the channel. In contrast, the term "transverse position" denotes position orthogonal to the

DETD mechanisms generally comprise any mechanisms in which a force acts directly on a particle(s) to position the particle(s) within a ***microfluidic*** network. Direct positioning mechanisms may be based on any suitable mechanism, including optical, electrical, magnetic, and/or gravity-based forces, among others. . . . particles. Suitable electrical mechanisms include "electrokinesis," that is, the application of voltage and/or current across some or all of a ***microfluidic*** network, which may, as mentioned above, move charged particles directly

(e.g., via electrophoresis) and/or indirectly, through movement of ions in

DETD mechanisms in which a force acts indirectly on a particle(s), for example, via fluid, to move the particle(s) within a ***microfluidic*** network longitudinally and/or transversely.

DETD [0199] Transverse positioning of particles and/or reagents in a ***microfluidic*** system may be mediated at least in part by a laminar flow based mechanism. Laminar flow based mechanisms generally comprise any positioning flow streams, preferably one flowing away from the junction. Due to the laminar flow properties of flow streams on a ***microfluidic*** scale, the unifying site may maintain the relative distribution of inlet flow streams after they unify as laminar outlet flow.

DETD [0203] Transverse positioning of particles and/or reagents in a ***microfluidic*** system may be mediated at least in part by a stochastic (or portioned flow) positioning mechanism. Stochastic transverse positioning mechanisms

DETD [0211] ***Microfluidic*** systems may include one or more retention mechanisms. A retention mechanism generally comprises any suitable mechanism for retaining (or holding, capturing, or trapping) particles at preselected positions or regions of ***microfluidic*** networks, including single or plural mechanisms, operating in series and/or in parallel. Retention mechanisms may act to overcome the positioning. .

DETD [0214] Retention mechanisms may be based at least partially on particle contact with any suitable physical barrier(s) disposed in a ***microfluidic*** network. Such particle-barrier contact generally restricts longitudinal particle movement along the direction of fluid flow, producing flow-assisted retention. Flow-assisted particle-barrier.

DETD [0218] Chemical interactions may be specific. Specific mechanisms may use specific ***binding*** pairs (SBPs), for example, with first and second SBP members disposed on particles and passage surfaces, respectively. Exemplary SBPs may below in Table 1, with the designations of first and second being arbitrary. SBP members may be disposed locally within ***microfluidic*** networks before, during and/or after formation of the networks. For example, surfaces of a substrate and/or a fluid layer component layer component are joined. Alternatively, or in addition, an SBP member may be locally associated with a portion of a ***microfluidic*** network after the network has been formed, for example, by local chemical reaction of the SBP member with the network (such as catalyzed by local illumination with light).

TABLE 1

Representative Specific ***Binding*** Pairs

First SBP Member	Second SBP Member
Antigen	antibody
Biotin	avidin or streptavidin
Carbohydrate	lectin or carbohydrate receptor
DNA	antisense DNA or DNA- ***binding*** protein
enzyme substrate or inhibitor	enzyme
Histidine	NTA (nitrilotriacetic acid)
IgG	protein A or protein G
RNA	antisense RNA

DETD Chemical interactions also may be relatively nonspecific. Nonspecific interaction mechanisms may rely on local differences in the surface chemistry of ***microfluidic*** networks. Such local differences may be created before, during and/or after passage/ ***microfluidic*** network formation, as described above. The local differences may result from localized chemical reactions, for example, to create hydrophobic or hydrophilic regions, and/or localized ***binding*** of materials. The bound materials may include poly-L-lysine, poly-D-lysine, polyethylenimine, albumin, gelatin, collagen, laminin, fibronectin, entactin, vitronectin, fibrillin, elastin, heparin

DETD force on the particles that is generally orthogonal to fluid flow. Such forces may be exerted by centrifugation of a

microfluidic chip and/or by particle movement within a fluid flow path (see Example 9). Magnetic force-based retention mechanisms may retain particles using magnetic fields, generated external and/or internal to a ***microfluidic*** system. The magnetic field may interact with ferromagnetic and/or paramagnetic portions of particles. For example, beads may be formed at

DETD a blind fill channel, where a channel has a inlet, but no outlet, either fixedly or transiently. For example, when the ***microfluidic*** ***device*** is made from a gas-permeable material such as PDMS, gas present in a dead-end channel can escape, or be

DETD ion(s), polymer(s), material(s), complex(es), mixture(s), aggregate(s), and/or biological particle(s), among others, that contacts a particle or particle population in a ***microfluidic*** system. Reagents may play a role in particle analysis, including operating as chemical/biological modulators (interaction reagents), detection/assay reagents, solvents, buffers

DETD modulators or biological modulators may include any reagent that is being tested for interaction with particles. Interaction generally includes specific ***binding*** to particles and/or any detectable genotypic and/or phenotypic effect on particles (or modulators). Further aspects of interactions and genotypic/phenotypic effects

DETD [0240] Particles in ***microfluidic*** systems may be exposed to physical modulators/conditions using non-fluid-mediated mechanisms. However, these "non-fluid-mediated" mechanisms may use properties of fluid to particles via fluid. The physical modulators/conditions may be applied to particles from sources that are external and/or internal to the ***microfluidic*** systems. Exemplary physical modulators/conditions may include thermal energy (heat), radiation (light), radiation (particle), an electric field, a magnetic field, pressure

DETD [0246] Particles manipulated by a ***microfluidic*** system may be analyzed by one or more measurement mechanisms at one or more measurement sites. The measurement mechanisms generally

DETD particle (or a component or derivative thereof) and its neighbors (e.g., other particles), the solvent (including any matrix) and/or the ***microfluidic*** system, among others, and may be used to characterize molecular size and/or shape, or to separate a sample into its

DETD mechanism may be used to detect particles and/or particle characteristics at any suitable detection site, internal and/or external to the ***microfluidic*** system.

DETD [0256] Suitable internal detection sites may include any site(s) in or on a ***microfluidic*** system (a chip). These sites may include channels, chambers, and/or traps, and portions thereof. Particles or particle characteristics may be

DETD [0257] Suitable external detection sites may include any site(s) away from or independent of a ***microfluidic*** system. External detection sites may be used to detect a particle or particle characteristic after removal of particles (or particle components) from a ***microfluidic*** system. These external sites may be used instead of and/or in addition to internal sites, allowing particles (or particle components) detected. These further manipulations and/or detection methods may overlap with, but preferably complement, the manipulations and/or methods performed in the ***microfluidic*** system, including mass spectrometry, electrophoresis, centrifugation, PCR, introduction into an organism, use in clinical treatment, and/or cell culture, among others

DETD localization, structure/modification, conformation, morphology, activity, number, and/or movement of DNA, RNA, protein, enzyme, lipid, carbohydrate, ions, metabolites, organelles, added reagent (***binding***), and/or complexes thereof, among others. The detected characteristics also may include cellular characteristics, such as any suitable cellular genotype or phenotype, including morphology, growth, apoptosis, necrosis, lysis, alive/dead, position in the ***cell*** cycle, activity of a signaling pathway, differentiation, transcriptional activity, substrate attachment, ***cell*** ***cell*** interaction, translational activity, replication activity, transformation, heat shock response, motility, spreading, membrane integrity, and/or neurite outgrowth, among others

DETD [0263] A ***microfluidic*** system may include any suitable number

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tion(s) between the cells and the modulator.
Moreover,. . .

DETD [0303] Genotypic assays may be conducted on cells in
microfluidic systems to measure the genetic constitution of
cells. The genotypic assays may be conducted on any suitable
cell or ***cell*** populations, for example, patient
samples, prenatal samples (such as embryonic, fetal, chorionic villi,
etc.), experimentally manipulated cells (such as transgenic
Nucleic acid hybridization with nucleic acids may be carried out with a
dye-labeled probe, a probe labeled with a specific ***binding***
pair (see Section VI), a stem-loop probe carrying an energy transfer
pair (such as a "molecular beacon"), and/or with a aspects may
include total DNA content (for example, 2N, 4N, 8N, etc., to measure
diploid, tetraploid, or polyploid genotypes and/or ***cell*** cycle
distribution), number or position of specific chromosomes, and/or
position of specific genes (such as adjacent the nuclear membrane,
another

DETD [0304] Phenotypic assays may be conducted to characterize cells in
microfluidic systems based on genetic makeup and/or
environmental influences, such as presence of modulators. These assays
may measure any molecular or

DETD [0305] Aspects of a whole ***cell*** or whole ***cell***
population may include number, ***size***, density, shape,
differentiation state, spreading, motility, translational activity,
transcriptional activity, mitotic activity, replicational activity,
transformation, status of one or more processes, intact/lysed,
live/dead, frequency/extent of apoptosis or necrosis,
presence/absence/efficiency of attachment to a substrate (or to a
passage), growth rate, ***cell*** cycle distribution, ability to
repair DNA, response to heat shock, nature and/or frequency of
cell ***cell*** contacts, etc.

DETD [0306] Aspects of ***cell*** organelles may include number,
size, shape, distribution, activity, etc. of a ***cell*** 's
(or ***cell*** population(s) nuclei, ***cell*** surface
membrane, lysosomes, mitochondria, Golgi apparatus, endoplasmic
reticulum, peroxisomes, nuclear membrane, endosomes, secretory granules,
cytoskeleton, axons, and/or neurites, among others.

DETD [0307] Aspects of ***cell*** constituents/components may include
presence/absence or level, localization, movement, activity,
modification, structure, etc. of any nucleic acid(s), polypeptide(s),
carbohydrate(s), lipid(s), ion(s), molecule, hormone,
metabolite, and/or a complex(es) thereof, among others. Presence/absence
or level may be measured relative to other cells or ***cell***
populations, for example, with and without modulator. Localization may
be relative to the whole ***cell*** or individual ***cell***
organelles or components. For example, localization may be cytoplasmic,
nuclear, membrane-associated, ***cell*** surface-associated,
extracellular, mitochondrial, endosomal, lysosomal, peroxisomal, and/or
so on. Exemplary cytoplasmic/nuclear localization may include
transcription factors that translocate between these two include
intracellular trafficking, such as protein targeting to specific
organelles, endocytosis, exocytosis, recycling, etc. Exemplary movements
may include endocytosis of ***cell*** surface receptors or
associated proteins (such as GPCRs, receptor tyrosine kinases, arrestin,
and/or the like), either constitutively or in response to ligand
binding. Activity may include functional or optical activity,
such as enzyme activity, fluorescence, and/or the like, for example,
mediated by kinases structure (such as association with
partners in, on, or about cells). Methods for measuring modifications
and/or structure may include specific ***binding*** agents (such as
antibodies, etc.), in vivo or in vitro incorporation of labeled
reagents, energy transfer measurements (such as FRET)

DETD are expressed from reporter genes), and/or the like. Methods
for measuring polypeptides may include enzymatic assays and/or use of
specific ***binding*** members (such as antibodies, lectins, etc.),
among others. Specific ***binding*** members are described in
Section VI.

DETD [0312] Interaction generally comprises any specific ***binding*** of
a modulator to a ***cell*** or population of cells, or any
detectable change in a ***cell*** characteristic in response to the
modulator. Specific ***binding*** is any ***binding*** that is
predominantly to a given partner(s) that is in, on, or about the
cell (s). Specific ***binding*** may have a ***binding***
coefficient with the given partner of about 10.sup.-3 M and lower, with

preferred specific ***binding*** coefficients of about 10 sup -4 M, 10 sup -6 M, or 10 sup -8 M and lower. Alternatively, interaction may be any change in a [illegible]

DETD [illegible] modulator may be labeled, such as with an optically detectable dye, and may be labeled secondarily after interaction with cells. ***Binding*** of the dye to the ***cell*** or cells thus may be quantified. Alternatively, or in addition, the ***cell*** may be treated or otherwise processed to enable measurement of a phenotypic characteristic produced by modulator contact, as detailed above.

DETD [0314] Cells and/or ***cell*** populations may be screened with libraries of modulators to identify interacting modulators and/or modulators with desired interaction capabilities, such as [illegible] effect (such as reporter gene response, change in expression level of a native gene/protein, electrophysiological effect, etc.) and/or coefficient of ***binding***. A library generally comprises a set of two or more members (modulators) that share a common characteristic, such as structure [illegible]

DETD [0316] ***Microfluidic*** assays of cells and/or populations may measure activity of signal transduction pathways. The activity may be measured relative to an [illegible]

DETD [0317] Signal transduction pathways generally comprise any flow of information in a ***cell***. In many cases, signal transduction pathways transfer extracellular information, in the form of a ligand(s) or other modulator(s), through the [illegible] an intracellular signal. The extracellular information may act, at least partially, by triggering events at or near the membrane by ***binding*** to a ***cell*** surface receptor, such as a G Protein Coupled Receptor (GPCR), a channel coupled receptor, a receptor tyrosine kinase, a receptor serine/threonine kinase, and/or [illegible] so on, which may result ultimately in altered gene expression. In other cases, modulators pass through the membrane and directly ***bind*** to intracellular receptors, for example with nuclear receptors (such as steroid receptors (GR, ER, PR, MR, etc.), retinoid receptors, retinoid [illegible]

DETD [illegible] based at least partially on the type of signal transduction pathway being assayed. Accordingly, signal transduction assays may measure ligand ***binding***, receptor internalization; changes in membrane currents; association of receptor with another factor, such as arrestin, a small G-like protein, such [illegible]

DETD [illegible] genotypic, phenotypic, and/or modulator interaction of cells and/or populations of cells. The cells and/or populations may be compared in distinct ***microfluidic*** systems or within the same ***microfluidic*** system. Comparison in the same ***microfluidic*** system may be conducted in parallel using a side-by-side configuration, as exemplified by Example 2, in parallel at isolated sites [illegible]

DETD [0322] ***Microfluidic*** systems may be used to perform single-cell assays, which generally comprise any assays that are preferably or necessarily performed on [illegible]

DETD [0324] ***Microfluidic*** systems may be used to sort or select single cells and/or ***cell*** populations. The sorted/selected cells or populations may be selected by stochastic mechanisms (see Example 2), ***size***, density, magnetic properties, ***cell*** surface properties (that is, ability to adhere to a substrate), growth and/or survival capabilities, and/or based on a measured characteristic of [illegible] specific phenotype, and/or the like). Cells and/or populations may be sorted more than once during manipulation and/or analysis in a ***microfluidic*** system. In particular, heterogeneous populations of cells, such as blood samples or clinical biopsies, partially transfected or differentiated ***cell*** populations, disaggregated tissues, natural samples, forensic samples, etc. may be sorted/selected. Additional aspects of ***cell*** sorting and suitable cells and ***cell*** populations are described above in Section III and below in Examples 9, 15, 22, and 26.

DETD [0326] ***Microfluidic*** systems may perform storage and/or maintenance functions for cells. Accordingly, cells may be introduced into ***microfluidic*** systems and cultured for prolonged periods of time, such as longer than one week, one month, three months, and/or one year. Using ***microfluidic*** systems for storage and/or maintenance of cells may consume smaller amounts of media and space, and may maintain cells in a more viable state than other storage/maintenance methods. Additional aspects of storing and maintaining cells in ***microfluidic*** systems are included in Section XI above and Example 10 below.

DETD [0328] ***Microfluidic*** systems may be used for any suitable virally based, organelle based, bead based, and/or vesicle based assays and/or methods. These assays may measure ***binding*** (or effects) of modulators (compounds, mixtures, polymers, biomolecules, cells, etc.) to one or more materials (compounds, polymers, mixtures, cells, etc.). . .

DETD [0331] The following examples describe selected aspects and embodiments of the invention, including methods of fabricating, integrating, and using ***microfluidic*** systems and devices and mechanisms for manipulation and analysis of particles. These examples are included for illustration and are not

DETD [0334] This example describes ***microfluidic*** systems for positioning and/or retaining single particles or groups of particles, based at least in part, on divergent flow paths.

DETD particles may be analyzed by any suitable method, including optical and electrical methods, among others. The described mechanisms use a ***microfluidic*** flow path that diverges to form a quasi-stagnant fluidic region at the position of divergence. Particles entering this quasi-stagnant fluidic region from a ***microfluidic*** stream experience a reduction in velocity, which may be exploited to effect their "soft landing" in a suitable retention structure. . .

DETD [0340] FIG. 2A shows a system 110 for ***microfluidic*** manipulation and/or analysis of particles, in accordance with aspects of the invention. System 10 includes (1) an input reservoir 112, (2) a ***microfluidic*** network 114 having three fluidic channels 116, 118, 120, and (3) two output or waste reservoirs 122, 124. Particles are. . .

DETD [0345] FIG. 2B shows another system 110' for ***microfluidic*** manipulation and/or analysis of particles, in accordance with aspects of the invention. The operational principles for system 110' of FIG. . .

DETD [0347] FIG. 3 shows yet another system 170 for ***microfluidic*** manipulation and/or analysis of particles, in accordance with aspects of the invention. System 170 includes (1) a fluidic network 172. . .

DETD this embodiment, gap 212 is slightly wider than the diameter of cells 206, so that it will accept only one ***cell***. In other embodiments, and/or for other cells, gap 212 may be wide enough to accept two or more cells. Whatever the width of gap 212, wall 190 and partitions 192, 194 form a retention site 214 at which ***cell*** 204, or cells may be stably retained. Once ***cell*** 204 is positioned at the retention site by trap 180, its presence may tend to block or diminish fluid flow. . . draw additional cells between partitions 192, 194. As a result, in some embodiments, trap 180 may preferentially retain only one ***cell***, automatically, without any need for optical monitoring during positioning and/or retention. Thus, retention site 214 may be dimensioned based on the ***size*** of cells to be retained. For example, eukaryotic cells typically are about 2 to 10 mu m in diameter, so gap. . .

DETD ***Microfluidic*** Systems for Trapping and Perfusing Particles

DETD [0350] This example describes ***microfluidic*** systems that position and retain single particles or sets of particles, and allow rapid, precise perfusion of the retained particles.

DETD a range of cell phenotypes or responses when the population is homogeneous or clonal. Therefore, studies of cells in a ***microfluidic*** environment would benefit from ***microfluidic*** systems that automatically position and/or retain a set of cells at a preselected site on a ***microfluidic*** chip. Furthermore, these studies would benefit from mechanisms that allow the retained set of cells to be perfused with selected. . .

DETD [0354] This example describes ***microfluidic*** systems that enable a user to trap multiple cells within a cell retention chamber, and perfuse the trapped cells with. . .

DETD [0356] FIGS. 5-11 show a system 250 for ***microfluidic*** analysis of cell populations. This system is described in detail below, including (a) system description, (b) system production, (c) system. . .

DETD [0358] FIG. 5 shows a portion of a system 250 for ***microfluidic*** analysis of cell populations. System 250 includes a ***microfluidic*** layer 252 and a control layer 254. ***Microfluidic*** layer 252 forms a ***microfluidic*** network 256 of interconnected channels depicted in blue and orange. Control layer 254 is positioned over and abutting the ***microfluidic*** layer, and includes valves and pumps (see also FIG. 8), depicted in purple. Exemplary dimensions and presented below for system 250. . .

DETD [0259] The ***microfluidic*** layer includes ***microfluidic*** channels with distinct geometries and functions. Blue, flow channels 258 have a semi-circular or arcuate cross-sectional profile and are positioned ... particle. Thus, these cell channels are used to position cells to preselected laminar flow streams and preselected regions of the ***microfluidic*** network. Perfusion channels 262, described more fully below, also are shown in orange and function to controllably perfuse retained cells.

DETD ... of reagent exposure to measure rapid cell responses may be conducted reproducibly with the rapid response times afforded by this ***microfluidic*** system.

DETD [0267] FIG. 8 shows additional aspects of ***microfluidic*** system 250. These additional aspects include macrofluidic reservoirs, and valves and/or pumps of the control layer that control fluid flow within the ***microfluidic*** network.

DETD ... the macroscopic world. Each reservoir or well functions as a fluidic inlet or outlet connected directly to at least one ***microfluidic*** channel. Fluidic inlet well A, shown at 330, provides for particle input, generally as a cell suspension. Fluidic inlet well B, shown ...

DETD ... 250 may be formed using any suitable method. In an exemplary approach, the system is formed by layering and fusing ***microfluidic*** layer 252, control layer 254, and a substrate layer, formed, for example, by a cover slip (not shown). Specifically, in this approach, the ***microfluidic*** and control layers are molded by soft lithography and then fused. Next, the resulting fused multilayer structure is bonded to the cover slip substrate layer. Finally, ***microfluidic*** channels are wetted with deionized water.

DETD [0285] The ***microfluidic*** system demonstrated here can be used for any suitable assay, such as screening compounds against a small population of cells, with the ***size*** of the small population be selected to be statistically representative of ***cell*** behavior. The particles may include cells and/or beads, among others. The cells may be nonadherent and/or adherent cells, either in suspension or attached to a substrate provided by the ***microfluidic*** system. The beads similarly may be nonadherent or adherent, and may be used to carry samples, reagents, and/or cells, among ...

DETD [0288] Communication passages 402 may be ***size***-selective channels configured to prevent movement of retained particles, generally cells, between each subsystem 250. However, passages 402 are configured to ... to, from, and/or between retained cells. Furthermore, perfusion mechanisms 268 may be used to determine the effect of reagents on ***cell*** ***cell*** communication mediated by passages 402.

DETD [0289] FIG. 11C shows a retention mechanism 410 that may be used in system 250 or any other suitable ***microfluidic*** system to form a positioned, two-dimensional array of retained particles. Mechanism 410 includes an array of individual traps 412 oriented ...

DETD ***Microfluidic*** Systems for Parallel Retention and/or Treatment of

DETD [0294] This example describes ***microfluidic*** mechanisms and systems that position a plurality of particles and/or reagents at discrete transverse regions and flow paths within a ...

DETD ... may need to be averaged over many experiments to achieve meaningful results. Therefore, it would be desirable to have a ***microfluidic*** system that positions, treats, and analyzes particles or groups of particles adjacent one another at a microscopic level, to allow ...

DETD [0398] The ***microfluidic*** systems described in this example position a plurality of particles or (particle populations) and/or reagents along distinct, transversely disposed flow ...

DETD [0401] The ***microfluidic*** systems of this example may allow more efficient and meaningful use of ***microfluidic*** space for comparative analysis of particles and/or reagents.

DETD [0404] FIGS. 12 and 13 show a ***microfluidic*** system 420 (Embodiment 1) for retaining separate populations of particles, and exposing the populations to one or more selected reagents.

DETD [0420] System 480 was tested as described below. ***Microfluidic*** chips were fabricated according to system 480 of FIG. 13A and used for analysis of flow patterns and particle treatment.

DETD [0435] FIG. 13L shows a portion of ***microfluidic*** system 1820 that may be used to separately address particles and/or reagents to sets

of particle traps. System 1820 includes alternative embodiments
traps 1826 are disposed in a transverse channel, such as channel 1798 or
a chamber, such as a ***cell*** chamber with ***size***
selective channel around its perimeter

DETD ***Microfluidic*** System for Multiplexed Analysis of Particles in an Array

DETD [0436] This example describes a ***microfluidic*** system that loads particles in a serially distributed set of particle retention sites, and separately addresses reagents to each of

DETD treating the cells in such microtiter plates, and measuring short-term consequences of such treatments, poses substantial technical hurdles. Therefore, a ***microfluidic*** system is needed that forms more reproducible arrays of individual cells or small groups of cells at distinct positions, and

DETD [0440] This example describes a ***microfluidic*** system that serially traps small sets of particles at preselected positions within the system, allowing treatment of the trapped particles of traps. Thus, this design may be used to integrate a large number of traps into a single system. This ***microfluidic*** system also reduces the number of control lines required, as single control lines regulate sets of fluidic channels, such as loading, but then fluidically isolated during particle treatment and measurement. This arrangement of the traps enables the fabrication of larger ***microfluidic*** systems that may be suitable for use in high throughput drug discovery. For example, system 510 has a footprint of 2

DETD [0441] FIG. 14 shows a ***microfluidic*** system 510 for forming and analyzing an array of particles. System 510 may be formed by any suitable technique, such as multilayer soft lithography, to include at least two distinct layers: (1) a ***microfluidic*** network layer 512, shown in blue and orange, and (2) a control layer 514, shown in pink. Channels having distinct

DETD [0442] ***Microfluidic*** layer 512 includes two orthogonally directed networks. Particle loading network 516 is used to input and position particles, so that

DETD ***Microfluidic*** ***Device*** for Forming and Analyzing a Particle Array Using a "Cell Comb"

DETD [0454] This example describes a ***microfluidic*** ***device*** for forming and analyzing arrays of small number of particles, such as cells; see FIGS. 17-20

DETD [0456] In many applications, it is necessary to form an array of ***cell*** analysis chambers, with each chamber containing the same number of cells. These chambers allow multiple experiments, such as drug screens, to be conducted in parallel, in a consistent and comparable fashion. Currently, standard analyses use wells of microtiter plates as ***cell*** chambers, distributing an equal volume of a ***cell*** suspension to each of the wells. The ***size*** of these chambers and thus the number of cells analyzed has been decreasing in response to efforts to reduce the lead to huge errors in the detected reaction signals. Accordingly, with even fewer cells per well, for example, with single ***cell*** assays or when cells of interest are in limited supply, microtiter plates do not provide an adequate ***cell*** analysis chamber unless cells are counted to place an equal number per well. Even then, microtiter plates are deficient for performing drug are difficult to measure with microtiter plates, because adding and mixing steps cannot be performed very rapidly. Therefore, many ***cell*** analyses would benefit from systems for efficiently loading, rapidly treating, and analyzing small numbers of cells.

DETD [0458] FIG. 17 shows a ***microfluidic*** ***device*** 610 for forming an array of single particles or small groups of particles. Device 610 includes an input channel 612

DETD all chambers 618. Alternatively, filter 622 may be formed by smaller "leak" channels within filter channel 616, or by posts ***obstacles*** or protrusions that extend into a portion of filter channel 616, or that are disposed adjoining or adjacent an end of the filter channel. The diameter of the smaller channels, or the spacing of the posts/ ***obstacles*** determines the size of particle retained in chamber 618. Thus, as long as the diameters of these smaller channels, or the maximum spacing between these posts/ ***obstacles*** are sufficiently less than the diameter of a particle to be retained, the particle will be confined to chamber 618. . .

DETD receive a fixed number of input particles 620, such as a single particle. Such input particles may have a common ***size*** , such as cells from a homogeneous ***cell*** population, or they may have a range of sizes, such as cells from blood. In some embodiments, the diameter of filter channel 616 allows ***size*** -selective retention of a single particle. For example, the diameter may be large enough to receive certain particles in a heterogeneous particle population, such as ***red*** ***blood*** ***cells*** , but small enough to exclude others, such as white blood cells. Filter 622 also acts ***size*** -selectively, as described above, so in combination with chamber 618, individual filter channels 616 may be designed to retain a single ***cell*** within a defined ***size*** range. Alternatively, individual filter channels may be designed to retain a group of two or more cells, with each ***cell*** having a minimum ***size*** that is retained by filter 622.

DETD [0468] ***Cell*** combs described in this example may be useful in a variety of applications. For example, ***cell*** combs may be useful in drug discovery, serving as replacements for microtiter plates in ***cell*** assays to provide tighter control of the ***cell*** numbers. With current technology, the fabrication of each ***cell*** chamber in a ***cell*** comb device can be carried out with precision. Therefore, ***cell*** assays may be performed with an array of cells formed using this device, with reduced signal variation from chamber to chamber, even with single ***cell*** assays. ***Cell*** combs may, more generally, be used with a variety of micron-sized particles, in addition to cells, such as fluorescently or enzymatically coated beads. This device also can operate in gas phase, as long as the ***size*** of the particles of interest is larger than the pore ***size*** of the filter units. ***Cell*** combs also can be cascaded so that objects of different sizes are filtered out at different stages.

DETD [0471] One goal of ***microfluidic*** systems is the capability of retaining particles at preselected positions for subsequent treatment and analysis. Traps that perform such retention

DETD been dimensioned to trap a single particle; however, they alternatively may be dimensioned to trap two or more particles. The ***microfluidic*** system with respect to which each retention mechanism is illustrated, particularly positioning mechanism 264 and perfusion mechanism 268, is described the positioning and perfusion mechanisms. However, the retention mechanisms presented in this example may be combined with any other suitable ***microfluidic*** mechanisms for particle analysis.

DETD [0475] FIG. 21 shows a ***microfluidic*** system 710 for positioning, retaining, and/or perfusing a single particle, in accordance with aspects of the invention. Portions of system

DETD [0477] FIG. 22 shows another ***microfluidic*** system 740 for positioning, retaining, and/or perfusing a single particle, in accordance with aspects of the invention. View 742 shows a color-coded schematic of a system 740, whereas view 744 shows a photograph of an actual ***microfluidic*** system formed according to view 742, but flipped horizontally. System 740 includes a trap 746 positioned centrally at T-junction 714.

DETD [0478] FIG. 23 shows yet another ***microfluidic*** system 790 for positioning, retaining, and/or perfusing a single particle, in accordance with aspects of the invention. System 790 includes 730 by about 5 .mu.m. View 802 shows a line representation of trap 792, but includes a portion 804 of ***microfluidic*** system outside of distal wall 716. Sectional views 806, 808 show how retention blocks 800 extend outward and downward from

DETD focus. In view 816, the focal plane is near the substrate surface, showing sharp lines at corners 820, where the ***microfluidic*** layer 822 contacts substrate 730. The bottom perimeter 824 of blocks 800 is blurry because bottom surface 814 is raised

DETD Mechanisms for Reusable ***Microfluidic*** Systems

DETD [0481] This example describes mechanisms that promote reuse of ***microfluidic*** systems, including mechanisms for release, collection, and/or resuspension of particles; see FIGS. 24-28.

DETD [0482] ***Microfluidic*** systems often are designed for single use. Such single use systems may be used to retain and analyze a single cell. macroscopic volumes of cells and reagents, and is time consuming for initialization. Thus, there is a need for a reusable

microfluidic system that releases retained particles after their analysis, freeing the system (or cells) for additional analysis.

DETD [0485] This example describes ***microfluidic*** mechanisms that enable formation of reusable ***microfluidic*** systems. These ***microfluidic*** mechanisms include (1) a particle release mechanism, (2) a particle collection mechanism, and (3) a particle suspension mechanism. The particle ... well over time. These three mechanisms alone, or in any suitable combination, may enable more efficient and economical use of ***microfluidic*** systems for particle analysis.

DETD [0487] FIG. 24 shows a ***microfluidic*** system 850 having a particle release mechanism 852 and a particle collection mechanism 854, in accordance with aspects of the ...

DETD [0491] Fluid flow through ***size*** -selective channel 872, and thus particle release, is controlled by valve V2 (see FIG. 24). Valve V2 is a control layer valve disposed over reservoir channel 870. When valve V2 is closed, reservoir channel is compressed, forcing fluid outward through ***size*** -selective channel 872 into trap 858. This releases trapped particles, propelling them out of trap 858 into a flow stream, such ... are closed, and the shield buffer is running. Thus, the main flow stream goes from the buffer wells to the ***cell*** culture area, described below. When valve V2 is opened, reservoir channel 870 expands, bringing fluid in through ***size*** -selective channel 868 and refilling the reservoir channel ...

DETD ... during release. Fluid flows through retention area 906 to outlet 910 by passing through filter channels 908, which act as ***size*** -selective channels that prevent released particles from flowing to the outlet. Thus, released particle are collected in retention area 906. When the collected particles are cells, the retention area may be used to culture cells to promote ***cell*** growth, differentiation, and/or response to a treatment, such as by perfusion mechanism 860. Alternatively, the retention area may be operatively ...

DETD [0499] Standard particle input mechanisms, such as inlet well 330 of FIG. 8, are sufficient for single use ***microfluidic*** systems. However, these mechanisms may be inadequate for reusable systems. In reusable systems, it may be desirable to load a ...

DETD [0500] FIG. 28 shows a particle suspension mechanism 920 that may be integrated into reusable ***microfluidic*** systems, such as systems 850 and 880 described above. This suspension mechanism helps to maintain particles in suspension and/or helps to resuspend settled particles during the course of analyses with a reusable ***microfluidic*** system. Mechanism 920 includes an inlet reservoir 922, recirculation channels 924, and pumping valves 926. Inlet reservoir 922 receives and.

DETD ***Microfluidic*** Mechanisms for Adjustable Reagent Delivery

DETD ... volumes of the reagent are dispensed to provide a range of doses. However, this approach may not be suitable with ***microfluidic*** systems, because it may not be practical to dispense metered volumes in a ***microfluidic*** system and because it may require a mixer to mix and thus dilute such a dispensed volume. Thus, there is a need for a ***microfluidic*** mechanism that dispenses a premixed reagent at a range of selected concentrations, using a small number of reagent stocks.

DETD ... and second reagents at a range of concentrations, in accordance with aspects of the invention. Dilution mechanism 960 includes a ***microfluidic*** layer 962 having first and second reagent reservoirs 964, 966, and first and second controllable flow channels 968, 970 acting ...

DETD [0512] The dilution mechanisms described above may be used as part(s) of any suitable ***microfluidic*** ***device*** for any suitable applications. For example, dilution mechanism 990 may be used in ***microfluidic*** system 250 in FIG. 8 of Example 2 to prepare and deliver a desired mixture of reagents for particle perfusion.

DETD ***Microfluidic*** Sorting Mechanisms Based on Centrifugal Forces

DETD [0515] ***Microfluidic*** analyses of particles may benefit from or even require sorting crude or heterogeneous input populations of particles into their components ... constantly monitored and actively directed to distinct sorting bins based on optical properties. Thus, there is a need for a ***microfluidic*** sorting mechanism that separates distinct particles, potentially passively, based on different physical properties of the distinct particles.'

DETD liquid columns having different heights in input and output reservoirs. However, these sorting mechanisms may be integrated into any suitable ***microfluidic*** system.

DETD [0519] FIGS. 31 and 32 show a ***microfluidic*** system 1020 having a sorting mechanism 1022 that separates particles according to physical differences between the particles, in accordance with may be formed with any suitable dimensions. Furthermore, mechanism 1022 may sort particles from any suitable source, such as a ***microfluidic*** treatment or analysis, into any desired number of outlet channels and/or other ***microfluidic*** mechanisms or structures, such as culture chambers, retention mechanisms, perfusion mechanisms, and/or the like.

DETD [0523] FIG. 33 shows a ***microfluidic*** system 1060 having a sorting mechanism 1062 with modified sorting parameters, in accordance with aspects of the invention. Sorting mechanism

DETD [0525] FIGS. 34 and 35 show another ***microfluidic*** system 1080 having a sorting mechanism 1082 with modified sorting parameters, in accordance with aspects of the invention. Sorting mechanism

DETD described in this example. The particles are split into two streams 1100 in the separation region. The lower stream is ***enriched*** for cells (red), and the upper stream is ***enriched*** for beads (green). Flow of particles through the system is powered by a 1 cm high column of fluid in the

DETD [0531] The systems shown in this example have the ability to passively ***enrich*** particles based on sorting mechanisms that distinguish physical properties of particles. The approximately two-fold ***enrichment*** obtained using these systems may be sufficient to facilitate or improve some ***microfluidic*** analyses. Furthermore, each of these systems may be modified and refined, and/or connected in series to improve ***enrichment*** of desired particles.

DETD ***Microfluidic*** Systems for Manipulating Sets of Particles

DETD [0532] This example describes ***microfluidic*** systems having relatively large chambers, in which larger sets of particles, such as adherent and/or nonadherent cells, can be retained.

DETD [0534] The introduction and/or removal of particles into and out of ***microfluidic*** systems at macroscopic/microscopic interfaces may inefficient and/or harmful. For introduction, particles must be placed in suspension and often are introduced example, by evaporation of inlet or outlet reservoir liquid. Accordingly, it is desirable to avoid repeatedly introducing and removing particles from ***microfluidic*** systems during a sequential set of assays. Therefore, there is a need for chambers for storing, treating, maintaining, measuring, and/or

DETD [0537] This example describes various ***microfluidic*** systems that address and solve some or all of the problems and issues cited above. These ***microfluidic*** systems may be formed using multilayer soft lithography, as described elsewhere in this Detailed Description and in the Cross References. Channels such molds may be used to form particle chambers of various dimensions. These channels and/or chambers may be integrated into ***microfluidic*** systems that include valves, pumps, rotary mixers, filters, sorters, multiplexers, perfusion mechanisms, and/or additional particle retention sites, among others, to

DETD [0539] FIGS. 39-43 illustrate exemplary ***microfluidic*** networks 1130 that include relatively large chambers 1132 for retaining particles, in accordance with aspects of the invention. These networks. result, this process allows formation of chambers with width to height ratios less than about 10:1 that do not collapse. In contrast, ***microfluidic*** channels having width to height ratios greater than 10:1 formed by a standard soft lithography process may collapse more frequently.

DETD posts may project downward from the roof of the channel to contact the substrate, generally being integrally formed in the ***microfluidic*** layer during fabrication of this layer. Alternatively, or in addition, these columns or posts may project upward from the substrate.

DETD [0544] FIG. 45 shows a ***microfluidic*** system 1180 having a ***microfluidic*** network 1130 through which fluid flow is more flexibly controlled. Specifically, fluid flow through chamber 1132 is controllable by two

DETD then allows the detached cells to be flushed from the system, either to output reservoir 1136 or to any additional ***microfluidic*** mechanism or set of mechanisms, as described

throughout this detailed description.

DETD [0547] FIG. 46 shows a ***microfluidic*** system 1210 with a cell chamber 1212 formed as a looped channel or ring structure, in accordance with aspects of

DETD [0549] FIGS. 47-49 shows another ***microfluidic*** system 1240 with a chamber 1242 formed as a looped channel or ring structure, in accordance with aspects of the

DETD [0557] FIG. 50A shows a system 1910 for depositing cells (or other particles) in a ***microfluidic*** chamber 1912, based on an asymmetrically disposed flow path. Particles and fluid flow into chamber 1912 from inlet channel 1914.

DETD [0565] FIG. 50C shows a ***cell*** chamber 1970 that may be used to deposit (and retain) cells in one or two compartments 1972, 1974. Compartments 1972, 1974 may be connected by radially arrayed ***size*** selective channels 1976 to form a "spoked wheel" structure. Cells (or other particles) may be inputted from first input channel 1978 and deposited in compartment 1972. Fluid may flow through ***size*** selective channels 1976 to second input channel 1980. Alternatively, or in addition, additional cells, such as a distinct ***cell*** type may be inputted from second input channel 1980 to be deposited in outer compartment 1974, with fluid flowing toward first input channel 1978. With each of the two compartments occupied by distinct ***cell*** populations, ***cell*** ***cell*** communication may be analyzed by passage of released ***cell*** components (or extended ***cell*** structures) through the ***size*** selective channels between the two compartments. In alternative embodiments, the first and second compartments may have any suitable geometry, such as increase the area of communication between the two compartments. Furthermore, additional compartments may be added to measure interactions between additional ***cell*** types.

DETD [0567] ***Cell*** chamber 1990 is a modified version of chamber 1970 that includes an overflow capability. Here, inner compartment 1972 acts as a chamber that is connected to overflow compartment 1992 by transverse passages 1994, in addition to ***size*** selective channels 1976. Accordingly, input channel 1978 may be used to direct most of inputted cells (or other particles) into inner

DETD [0569] The ***microfluidic*** systems described here may be used for the manipulation of adherent and nonadherent cells. For example, after introduction to a

DETD concentration has been achieved, cells are loaded using a manual pipettor into the input well and cells flow into the ***microfluidic*** channel structures under the head flow generated by the column of liquid. Once adhered, adherent cells can be resuspended in the ***microfluidic*** channel by addition of trypsin-EDTA or other cell detaching agents.

DETD [0571] The ***microfluidic*** layer and substrate may be treated (or left untreated) to promote cell flow, cell viability, cell adhesion or nonadhesion, cell

DETD Systems for Electrophysiological Analysis of Cells in a ***Microfluidic*** Environment

DETD [0572] This example describes ***microfluidic*** systems for positioning, retaining, treating, and/or measuring cells, particularly for electrophysiological analyses; see FIGS. 51-58.

DETD approach, a glass electrode with a diameter of about 0.1-1 .mu.m is electrically sealed against the membrane of a single ***cell*** surrounding a membrane "patch" on the ***cell*** . The patch then may be left intact, separated from the ***cell*** "perforated" with channel forming agents, or penetrated, based on the type of information desired. With both intact patches and patches separated from a ***cell*** , the ***size*** of the patch and the density of channels in the membrane determine the number of channels being analyzed. Thus, different of channels in "macropatch recording." Alternatively, membrane patches can be perforated or penetrated to measure electrical properties of the entire ***cell*** membrane, in "whole ***cell*** " patch clamp studies. Perforated patches introduce a channel forming agent, such as the antibiotics nystatin or amphotericin B, into the membrane. Perforated patches enable whole ***cell*** recording of channel activity with loss of larger cytoplasmic components. Penetrated patches place an electrode inside a ***cell*** so that the electrode and the ***cell*** 's cytoplasm are continuous. Accordingly, penetrated patches also enable whole- ***cell*** patch-clamp recording.

DETD [0578] This example describes ***microfluidic*** devices that allow measurements of ion channel activity. These devices position a single cell in abutment with an aperture, so

DETD [0580] FIG. 51 shows a ***microfluidic*** ***device*** 1310 for measuring ion currents, in accordance with aspects of the invention. Device 1310 includes a planar patch clamp electrode

DETD elsewhere in this Detailed Description. The fluidic layer may be controlled by any suitable control mechanism, such as an overlying ***microfluidic*** control layer 1320. The base layer may be formed out of any suitable material, such as glass, plastic, and/or an

DETD [0584] FIGS. 52-58 shows a ***microfluidic*** system 1340 for single cell patch clamp recordings, in accordance with aspects of the invention. System 1340 includes a fluid layer network 1342

DETD [0588] Cell positioning mechanism 1354 generally comprises any mechanism that acts to position single cells within ***microfluidic*** network 1342. In addition to simple flow channels, the cell positioning mechanism may include a focusing mechanism 1360. Focusing mechanism 1360

DETD [0589] ***Cell*** positioning mechanism 1354 stochastically segregates single cells using a divided flow mechanism 1374, downstream from focusing mechanism 1360 or 1372; see 1380, 1382 (labeled "W1" and "W2," respectively, in FIG. 54). These outlet channels include a narrowed portion 1384 and a ***size*** restrictive channel 1386 that determine the relative flow rate through each corresponding outlet channel. Narrowed portion 1384 has a substantially larger diameter than ***size*** selective channel 1386, so that most of the flowing fluid (and cells) passes through narrowed portion 1384. However, some fluid passes through ***size*** restrictive channel 1386, eventually bringing a single ***cell*** 1388 to the mouth of the channel

DETD [0590] ***Cell*** retention mechanism 1356 generally comprises any mechanism for retaining a ***cell*** at a desired position, generally adjacent an orifice and/or electrode(s). Here, the ***cell*** retention mechanism functions at the channel mouth; see FIGS. 54 and 57. In particular, ***cell*** 1388 cannot enter ***size*** restrictive channel 1386 because the ***cell*** is too large. However, the pressure drop across ***size*** restrictive channel 1386 pulls ***cell*** 1388 against the channel mouth, holding ***cell*** 1388 in position. Positioned ***cell*** 1388 may restrict or block flow through ***size*** restrictive channel 1386, so that additional cells no longer are urged toward channel 1386. ***Cell*** 1388 also is positioned over an orifice 1390 (see FIG. 56) defined by the substrate layer. In alternative embodiments, single. .

DETD [0595] ***Microfluidic*** system 1340 may be configured in many suitable ways. For example, reagent inlet channels may unite, entering chamber E1 through

DETD ***Microfluidic*** System for Multiplexed Analysis of Cells by Patch Clamp

DETD [0596] This example describes ***microfluidic*** systems for performing electrophysiological analysis on one or more cells out of a set of single cells; see FIGS. 59-61

DETD [0601] This example provides a multiplexed version of a single aperture ***microfluidic*** ***device*** with a defined number ("n") of individually controllable apertures. Each individually controllable aperture may be used to analyze a single

DETD [0606] FIG. 60 shows a ***microfluidic*** ***device*** 1450 that is a multiplexed version of device 1420, in accordance with aspects of the invention. Device 1450 may include

DETD [0608] Device 1450 may be modified in any suitable fashion incorporating any suitable ***microfluidic*** mechanisms, such as those described in this Detailed Description. For example, device 1450 may be structured to load cells serially, described above in Examples 2-5. Furthermore, device 1450 may be included in an array of such devices to form a ***microfluidic*** array. Alternatively, or in addition, device 1450 may include a perfusion mechanism, such as that described in Examples 2 and

DETD Multilayer Mold Fabrication Method of Varying Height and/or Cross-Sectional Geometries of Molded ***Microfluidic*** Structures

DETD [0610] This example describes a method for producing, by soft lithography, ***microfluidic*** devices in which the cross-sectional geometry and/or height of structures within and/or between ***microfluidic*** networks vary; see FIGS. 62-71.

DETD [0613] A ***microfluidic*** network may include structures having a variety of functions. For example, regulatable channels may include deflectable valves, acting to partially . . . of the width of the channels, rather than just the central portion, as with arcuate channels. Other channels may be ***size*** selective or particle restrictive, preventing entry of particles greater than a given ***size***. These particle restrictive channels may have a height that is less than the diameter of particles of interest. Furthermore, ***microfluidic*** networks may include ***cell*** /culture chambers with roof heights that are greater than more narrow channels, as described in Example 10, to improve the functionality . . .

DETD . . . be heated to round their edges into an rounded/arcuate configuration. Accordingly, these remaining regions of the resulting mold may produce ***microfluidic*** channels of complementary structure using soft lithography. In other embodiments, multiple layers of photoresist may be built up by sequential . . .

DETD [0614] Despite the importance of varying height and/or cross sectional shape across a ***microfluidic*** network, molds formed from a single layer of selectively removable material, such as photoresist, may not allow sufficient flexibility in the structure of a ***microfluidic*** network formed from the mold. For example, the depth to which the single layer may be removed cannot be varied . . .

DETD . . . in this example may be used to form channels with different cross sectional geometries and/or heights at distinct positions within a ***microfluidic*** network. A mold is fabricated using plural layers of photoresist that are each individually patterned, selectively removed according to the . . . pattern is the sum of the remaining portions from each of the plural layers. Using the mold to form a ***microfluidic*** network allows various types of channels or other passages to be formed. Channels with a rounded/arcuate cross sectional shape may be . . . to promote particle movement and to enable precise delivery of one or more particles to a specific area of a ***microfluidic*** network. The specific area can be as small as the dimension of a single particle, such as a cell. These structures and other suitable ***microfluidic*** structures may be produced using the method described below. This method focuses on formation of a fluid layer, but may be suitable for any portion(s) of a ***microfluidic*** system, including a control layer or a base layer (see Example 11).

DETD . . . The fluid layer mold may be used subsequently in a second series of steps, as described below, to mold a complementary ***microfluidic*** layer by soft lithography. FIGS. 62-68 illustrate how fluid layer mold 1480 may be formed by sequentially disposing, patterning, and selectively . . .

DETD [0627] The fluid layer and control layer molds fabricated above may be used to mold a ***microfluidic*** chip using any suitable material, particularly an elastomeric material, such as polydimethylsiloxane (PDMS). Exemplary PDMS elastomers are General Electric Silicones . . .

DETD [0631] FIGS. 70 and 71 show photographic images of fluid layer molds and the corresponding ***microfluidic*** chips formed with these molds. The ***microfluidic*** networks represented here, have been shown and described in system 1340 of Example 11 (FIG. 70) and in a modified.

DETD Detection System for Kinetic Analyses in ***Microfluidic*** Systems

DETD . . . detection system, including a modulation-demodulation method and the use of tracer materials, for analysis of kinetic reactions involving particles in ***microfluidic*** systems; see FIGS. 71A-F.

DETD [0634] ***Microfluidic*** systems may be used to measure the kinetics of many aspects of cellular metabolism. However, metabolic processes of physiological significance . . .

DETD . . . a result, some cellular processes that occur over relatively longer time periods may be more difficult to monitor in a ***microfluidic*** system due to this photobleaching . . .

DETD . . . source produces a reduced emission signal and signal to noise ratio, since the emission signal is proportional to the illumination intensity. Therefore, ***microfluidic*** analyses would benefit from a detection system that reduces photobleaching, increases the ratio of signal to noise, and/or allows kinetic analysis of . . .

DETD [0638] This example describes an exemplary detection system for use with ***microfluidic*** assays, in accordance with aspects of the invention. The detection system may include a modulation-demodulation mechanism, see FIGS. 71B-71E. This . . .

DETD . . . include a conventional microscope or other suitable optical

or partially or wholly integrated with, a

a Modulation-Demodulation Mechanism for

experiment.

Example 2. Dye-labeled streptavidin (reagent) is exposed to the.

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Microfluidic Analysis of a Heterogeneous Particle

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Microfluidic Interaction of Specific ***Binding*** Pairs

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T-cells were cultured in RPMI. Acridine Orange was

scope. The microscope's 63.times. oil-immersion lens

nd Detecting Cell Apoptosis in a ***Microfluidic***

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exposing PS to the ***cell*** exterior..sup.6 In leukocyte

apoptosis, PS on the outer surface of the ***cell*** marks the ***cell*** for recognition and phagocytosis by macrophages. sup.7,8 The human anticoagulant, annexin V, is a 35-36 kD Ca sup.2 dependent, phospholipid ***binding*** protein that has a high affinity for PS. sup.9 Annexin V can identify apoptotic cells by ***binding*** to PS exposed on the outer leaflet. sup.10 Bound annexin V may be detected through a dye, a specific ***binding*** member conjugated to annexin V, an anti-annexin V antibody, and/or the like.

DETD [0774] This example demonstrates induction and detection of cell apoptosis in a ***microfluidic*** system. Jurkat cells are positioned and retained in a ***microfluidic*** system, and then programmed cell death is initiated by exposure of these cells to hydrogen peroxide. Translocation of PS to ...

DETD [0775] Materials used were as follows. ***Microfluidic*** chips were constructed based on system 250 of Example 2. Jurkat T-cells were cultured in RPMI. The VYBRANT Apoptosis Assay ...

DETD [0781] ***Binding*** Buffer (BB) was loaded into the shield buffer well of the chip ...

DETD Analysis of Aquatic Microorganisms in a ***Microfluidic*** System

DETD [0816] This example describes the capture and visualization of aquatic microorganisms, such as plankton, using a ***microfluidic*** system.

DETD [0820] This example provides a ***microfluidic*** system capable of manipulating and detecting small plankton, particularly picoplankton (0.2 .mu.m), ultraplankton (2-5 .mu.m), and/or nannoplankton (5-60 .mu.m). Plankton may be retained, treated, and/or detected in an integrated ***microfluidic*** environment.

DETD [0821] Plankton were manipulated and detected in a ***microfluidic*** system as follows. A sample of seawater was collected from San Francisco Bay and centrifuged to concentrate organisms in the sample. A 20 .mu.L aliquot of the concentrated sample was loaded into the input reservoir of ***microfluidic*** system 250, described in Example 2 above. Naturally fluorescent plankton were retained in chamber 270 and detected successfully by fluorescent microscopy.

DETD ... example, plankton may be collected from freshwater sources or cultured, an aqueous plankton sample may be loaded directly into a ***microfluidic*** environment without concentration, and/or retained plankton may be exposed to any suitable reagents. Alternatively, or in addition, ***microfluidic*** systems may be used that sort a heterogeneous population of plankton according to a physical property (such as size or density, among others) or a measured property/characteristic (such as labeling with a dye and/or specific ***binding*** member).

DETD Analysis of Membrane Trafficking in a ***Microfluidic*** System using Membrane Dyes

DETD [0823] This example describes ***microfluidic*** analysis of membrane trafficking pathways in cells treated with membrane-labeling dyes.

DETD [0826] Some "FM" dyes available from Molecular Probes ***bind*** to ***cell*** membranes. Thus these FM membrane dyes may be used as general purpose probes for endocytosis, because they are generally nontoxic. FM ...

DETD ... of these dyes. These two FM dyes have substantially nonoverlapping emission spectra. II) Test the affinity of FM dyes for ***microfluidic*** chips formed with PDMS, to define a background level of staining. III) Trap a Jurkat cell in a ***microfluidic*** chip and perform two-color staining of the cell using the two FM membrane dyes.

DETD [0829] Materials used for this analysis included the following. FM 1-43 and FM 4-64 were obtained from Molecular Probes. ***Microfluidic*** chips were produced based on system 250 of Example 2. Results were collected and recording using an inverted fluorescent microscope.

DETD [0838] Labeling of the ***microfluidic*** chip with the FM membrane dyes to determine background signal was carried out as follows.

DETD [0845] Cells were labeled with FM dyes in a ***microfluidic*** system as follows.

DETD [0846] Unlabeled Jurkat cells were loaded and captured in a ***microfluidic*** chip using PBS as a carrier buffer.

DETD [0858] Protocol B produced significant background labeling of ***microfluidic*** chips formed with PDMS, using either dye. The PDMS may be surface-modified to minimize ***binding*** of these dyes to the chip.

DETD [0859] Protocol C was foiled by the high background produced by dye

binding to PDMS. After trapping a single ***cell*** in the chip, FM 1-43 bound to the chip more efficiently than to the membrane of the trapped ***cell***.

DETD Capturing Cells in Single-Cell or Multi-Cell ***Microfluidic*** Chambers

DETD [0860] This example describes capture of a single cell or a cell population in a ***microfluidic*** system; see FIGS. 80-82.

DETD Fixing and Staining Cells in a ***Microfluidic*** System

DETD [0862] This example describes the use of a ***microfluidic*** system to fix a cell with an organic solvent, methanol, and label the cell with acridine orange; see FIG. 83.

DETD ***Microfluidic*** Mechanism for Measuring Cell Secretion

DETD [0865] This example describes the structure and use of a soft lithography based ***microfluidic*** system for measuring secretion of molecules, complexes, and/or small particles from cells.

DETD [0866] Many ***cell*** analyses measure release and/or secretion of materials from cells. In some cases, the cells secrete material naturally. For example, neurons cells for secretion of endocrine hormones, such as insulin, growth hormone, prolactin, steroid hormones, etc., and a broad range of ***cell*** types for secretion of cytokines. In other cases, cells are lysed to define an aspect of their internal contents. However, solution. Accordingly, such secreted or released materials may be difficult to analyze without concentrating them and/or without using immobilized, high-affinity ***binding*** partners, for example, in ELISA.

DETD [0867] ***Microfluidic*** systems may ameliorate some of the difficulties associated with measuring material released from cells, but may introduce additional considerations. In ***microfluidic*** systems, cells may be grown in isolated chambers having small volumes, as described above in Example 10. The chambers may However, to maintain the released materials in a concentrated form, the chambers may be isolated from other portions of the ***microfluidic*** network. Such isolated chambers do not promote ready analysis of the released materials, since the materials may be isolated from analytical reagents and may be difficult to collect without substantially diluting the released materials. Therefore, a ***microfluidic*** mechanism is needed that allows material released from cells to be collected and/or analyzed in a distinct fluidic compartment that is not part of a primary fluidic layer of a ***microfluidic*** system.

DETD [0868] This example provides a ***microfluidic*** system having a cell chamber and a separate material collection compartment that communicate fluidically through a semi-permeable membrane. The semi-permeable in another compartment of the fluid layer, a compartment above the fluid layer, and/or below the substrate. For example, the ***microfluidic*** system may include a layer similar to the base layer of Example 11.

DETD ***Microfluidic*** Analysis of a Heterogeneous Particle Population, Part II

DETD [0869] This example describes ***microfluidic*** systems for sorting and analyzing heterogeneous populations of particles, such as blood samples, based on differences in particle size; see

DETD [0871] This example provides a ***microfluidic*** system 1650 that selectively retains and analyzes larger particles from a mixture of larger and smaller particles; see FIGS. 84

DETD may functionally interconnect as follows. Input mechanism 1652 introduces particles from a particle sample placed in particle input reservoir 1666 into ***microfluidic*** network 1668 of system 1650. Particles are moved by positioning mechanism 1654 to filtration mechanism 1656 by flow along inlet

DETD [0878] System 1650 first separates white blood cells from smaller ***red*** ***blood*** ***cells*** and platelets. These separated white blood cells are directed to a retention site, retained, and then processed by the perfusion

DETD set of particle-selective channels or chamber channels 1704 also may be disposed around the perimeter of capture chamber 1706. Accordingly, ***red*** ***blood*** ***cells*** may travel to flow through chambers 1708 and then waste reservoirs 1692, 1694 along a substantial area formed by inlet channel 1702 and chamber 1706. In particular, travel of ***red*** ***blood*** ***cells*** through particle-selective channels 1700 from inlet channel 1702 may avoid clogging chamber channels 1704. However, the white blood cells may. . .

DETD Then, valve V2 may be opened to allow the carrying buffer provided by alternative input mechanism 1678 to wash residual ***red*** ***blood*** ***cells*** out of chamber 1706. At this point, waste reservoirs 1692, 1694 may be emptied to avoid reverse flow of the ***red*** ***blood*** ***cells*** back into chamber 1706.

DETD . . . plan view of a perfusion device for exposing particles to an array of different reagents or different reagent concentrations. Here, ***microfluidic*** passage device 2000 provides a plurality of growth/perfusion chambers 2020 for loading particles, such as cells, through loading passage 2010. . .

DETD . . . 24 depict a top plan view of a device being used to measure the response of cells to a chemo-attractant. ***Microfluidic*** passage device 2200 provides reagent loading chamber 2330, wherein reagent is metered into reagent chamber 2300 by the opening of. . .

CLM What is claimed is:
1. A ***microfluidic*** ***device*** for treating a particle comprising: (a) an input mechanism for introducing a fluid sample containing a particle; (b) a ***microfluidic*** passage in fluid communication with said input mechanism; (c) a positioning mechanism in fluid communication with said ***microfluidic*** passage, said positioning mechanism for positioning said particle in said ***microfluidic*** passage while contained in said fluid sample; (d) a retention mechanism for retaining said particle upon being positioned by said. . .

CLM What is claimed is:
2. The ***microfluidic*** ***device*** of claim 1 further comprising a release mechanism for releasing said particle from said retention mechanism.

CLM What is claimed is:
3. The ***microfluidic*** ***device*** of claim 2 further comprising an output mechanism for outputting said particle from said ***microfluidic*** ***device*** .

CLM What is claimed is:
4. The ***microfluidic*** ***device*** of claim 2 further comprising a cell culture mechanism for culturing said particle.

CLM What is claimed is:
5. The ***microfluidic*** ***device*** of claim 1 further comprising a control mechanism for determining aspects of the flow rate or path of the sample. . .

CLM What is claimed is:
6. The ***microfluidic*** ***device*** of claim 5, wherein said control mechanism is a valve in communication with said ***microfluidic*** passage.

CLM What is claimed is:
7. The ***microfluidic*** ***device*** of claim 6, wherein said ***microfluidic*** ***device*** is formed from a multi-layer elastomeric block and wherein said valve is formed from an elastomeric membrane within said elastomeric. . .

CLM What is claimed is:
8. The ***microfluidic*** ***device*** of claim 6, wherein said control mechanism is a pump in communication with said ***microfluidic*** passage.

CLM What is claimed is:
9. The ***microfluidic*** ***device*** of claim 8, wherein said ***microfluidic*** ***device*** is formed from a multi-layer elastomeric block and wherein said pump is formed from an elastomeric membrane within said elastomeric. . .

CLM What is claimed is:
10. The ***microfluidic*** ***device*** of claim 1, wherein said ***microfluidic*** ***device*** comprises a multi-layered elastomeric block having a control layer having an elastomeric membrane deflectable into said ***microfluidic*** passage in a fluidic layer to determine the flow rate or path of a fluid in said ***microfluidic*** passage.

CLM What is claimed is:

CLM What is claimed is:
11. The ***microfluidic*** ***device*** of claim 1, wherein said ***microfluidic*** ***device*** comprises a layer including a material selected from the group consisting of elastomers, polydimethylsiloxane, plastic, polystyrene, polypropylene, polycarbonate, glass, ceramic,. . .

CLM What is claimed is:
12. The ***microfluidic*** ***device*** of claim 1, wherein said ***microfluidic*** passage has is less than about 500 micrometers wide.

CLM What is claimed is:
13. The ***microfluidic*** ***device*** of claim 1, wherein said ***microfluidic*** passage further comprises an adjacent passage joining said ***microfluidic*** passage at a junction or branch, said adjacent passage being selected from the group consisting of inlet passage, outlet passage,. . .

CLM What is claimed is:
14. The ***microfluidic*** ***device*** of claim 13, wherein said adjacent passage is a dead-end passage.

CLM What is claimed is:
15. The ***microfluidic*** ***device*** of claim 13 further comprising said adjacent passage manipulating said particle.

CLM What is claimed is:
16. The ***microfluidic*** ***device*** of claim 15, wherein said particle manipulating is selected from the group of positioning, sorting, retaining, treating, detecting, propagating, storing,. . .

CLM What is claimed is:
17. The ***microfluidic*** ***device*** of claim 1, wherein said particle is selected from the group consisting of cells, eukaryotic cells, prokaryotic cells, plant cells,. . .

CLM What is claimed is:
18. The ***microfluidic*** ***device*** of claim 17, wherein said particle is a plurality or an aggregate of particles.

CLM What is claimed is:
19. The ***microfluidic*** ***device*** of claim 18, wherein said plurality of particles is a complex mixture containing different particles.

CLM What is claimed is:
20. The ***microfluidic*** ***device*** of claim 19, wherein said complex mixture containing different particles is whole blood or serum or bodily fluid.

CLM What is claimed is:
21. The ***microfluidic*** ***device*** of claim 1, wherein said particle is an egg or embryo.

CLM What is claimed is:
22. The ***microfluidic*** ***device*** of claim 1, wherein the input mechanism is a receptacle or well in fluid communication with said ***microfluidic*** passage.

CLM What is claimed is:
23. The ***microfluidic*** ***device*** of claim 22, wherein the input mechanism has a volume greater than a volume defined by said ***microfluidic*** passage.

CLM What is claimed is:
24. The ***microfluidic*** ***device*** of claim 1 further comprising a facilitating mechanism in communication with or acting upon said input mechanism.

CLM What is claimed is:
25. The ***microfluidic*** ***device*** of claim 24, wherein said facilitating mechanism is selected from the group consisting of gravity, fluid pressure, centrifugal pressure, pump. . .

CLM What is claimed is:
26. The ***microfluidic*** ***device*** of claim 1, wherein said positioning mechanism is a direct positioning mechanism or an indirect

positioning mechanism.

CLM What is claimed is:
27. The ***microfluidic*** ***device*** of claim 26, wherein said direct positioning mechanism is a force selected from the group consisting of optical, electrical, magnetic,. . .

CLM What is claimed is:
28. The ***microfluidic*** ***device*** of claim 27, wherein said electrical force is selected from the group consisting of electrophoretic, electroosmotic, electroendoosmotic, and dielectrophoretic.

CLM What is claimed is:
29. The ***microfluidic*** ***device*** of claim 26, wherein said indirect positioning mechanism is a longitudinal indirect positioning mechanism or a transverse indirect positioning mechanism.

CLM What is claimed is:
30. The ***microfluidic*** ***device*** of claim 29, wherein said indirect positioning mechanism is facilitated by a pump or a valve associated with said ***microfluidic*** ***device*** .

CLM What is claimed is:
31. The ***microfluidic*** ***device*** of claim 29, wherein said transverse indirect positioning mechanism is facilitated by a fluid flow stream at a passage junction,. . .

CLM What is claimed is:
32. The ***microfluidic*** ***device*** of claim 31, wherein said passage junction is unifying or dividing.

CLM What is claimed is:
33. The ***microfluidic*** ***device*** of claim 29, wherein said transverse indirect positioning mechanism is a laminar flow-based transverse positioning means.

CLM What is claimed is:
34. The ***microfluidic*** ***device*** of claim 29, wherein said transverse indirect positioning mechanism is a stochastic transverse positioning mechanism.

CLM What is claimed is:
35. The ***microfluidic*** ***device*** of claim 34, wherein said stochastic transverse positioning mechanism randomly selects said particle from a population of particles by lateral. . .

CLM What is claimed is:
36. The ***microfluidic*** ***device*** of claim 29, wherein said transverse indirect positioning mechanism is a centrifugal forced-based transverse positioning mechanism.

CLM What is claimed is:
37. The ***microfluidic*** ***device*** of claim 1 wherein said retention mechanism selectively retains said particle at a pre-selected region within said ***microfluidic*** ***device*** .

CLM What is claimed is:
38. The ***microfluidic*** ***device*** of claim 37, wherein said retention mechanism retains said particle by overcoming or counteracting a force caused by said positioning. . .

CLM What is claimed is:
39. The ***microfluidic*** ***device*** of claim 1, wherein said retention mechanism is a trap or barrier-based retention mechanism.

CLM What is claimed is:
40. The ***microfluidic*** ***device*** of claim 39, wherein said barrier based retention mechanism is restricts longitudinal movement of said particle in or adjacent said ***microfluidic*** passage.

CLM What is claimed is:
41. The ***microfluidic*** ***device*** of claim 38, wherein said retention mechanism is a protrusion extending, fixedly or transiently, into or adjacent said ***microfluidic*** passage to

restrict longitudinal movement of said particle.

CLM What is claimed is:
42. The ***microfluidic*** ***device*** of claim 36, wherein said direct positioning mechanism is a chemical retention mechanism.

CLM What is claimed is:
43. The ***microfluidic*** ***device*** of claim 42, wherein said chemical retention mechanism is based on a specific affinity between said particle and said retention. . . .

CLM What is claimed is:
44. The ***microfluidic*** ***device*** of claim 1, wherein said treatment mechanism is a fluid-mediated mechanism or a non-fluid mediated mechanism.

CLM What is claimed is:
45. The ***microfluidic*** ***device*** of claim 1, wherein said treatment mechanism exposes said particle to a reagent or physical condition.

CLM What is claimed is:
46. The ***microfluidic*** ***device*** of claim 45, wherein said reagent is selected from the group consisting of chemical modulator, biological modulator, agonist, antagonist, hormone,. . .

CLM What is claimed is:
47. The ***microfluidic*** ***device*** of claim 46, wherein said reagent attracts or repels said particles.

CLM What is claimed is:
48. The ***microfluidic*** ***device*** of claim 45, wherein said reagent induces or inhibits cell particle proliferation.

CLM What is claimed is:
49. The ***microfluidic*** ***device*** of claim 45, wherein said reagent is cytotoxic.

CLM What is claimed is:
50. The ***microfluidic*** ***device*** of claim 44, wherein said fluid mediated mechanism further comprises a fluid treatment and wherein said particles are introduced to said. . .

CLM What is claimed is:
51. The ***microfluidic*** ***device*** of claim 44, wherein said fluid mediated mechanism functions in conjunction with the functioning of said positioning mechanism.

CLM What is claimed is:
52. The ***microfluidic*** ***device*** of claim 51, wherein said positioning mechanism is a transverse positioning mechanism for moving said particle into and out of. . .

CLM What is claimed is:
53. The ***microfluidic*** ***device*** of claim 45, wherein said physical condition is selected from the group consisting of heat, light, radiation, sub-atomic particles, electric. . .

CLM What is claimed is:
54. The ***microfluidic*** ***device*** of claim 1, wherein said measurement mechanism is a detector associated with said ***microfluidic*** ***device*** that detects a characteristic of said particle or caused by said particle.

CLM What is claimed is:
55. The ***microfluidic*** ***device*** of claim 54, wherein said detector is selected from the group consisting of spectroscopes, electronic sensors, hydrodynamic sensors, imaging systems,. . .

CLM What is claimed is:
56. The ***microfluidic*** ***device*** of claim 54, wherein said detector detects multiple values.

CLM What is claimed is:
57. The ***microfluidic*** ***device*** of claim 54, wherein said detector employs a detection mode that is selected from the group consisting of time independent, time-dependent,. . .

CLM What is claimed is:

58. The ***microfluidic*** ***device*** of claim 54, wherein said detector is a spectroscopic detector that detects a signal produced of a type selected from. . .

CLM What is claimed is:

59. The ***microfluidic*** ***device*** of claim 54, wherein said detector is an electrical detector capable of detecting a signal selected from the group consisting. . .

CLM What is claimed is:

60. The ***microfluidic*** ***device*** of claim 54, wherein said detector is a hydrodynamic detector which detects a hydrodynamic interaction between said particle and a fluid, another particle, or said ***microfluidic*** passage.

CLM What is claimed is:

61. The ***microfluidic*** ***device*** of claim 60, wherein said interaction included a hydrodynamic interaction selected from the group consisting of chromatography, sedimentation, viscometry, electrophoresis.

CLM What is claimed is:

62. The ***microfluidic*** ***device*** of claim 54, wherein said detector is an imaging detector for creating and analyzing images of said particle(s).

CLM What is claimed is:

63. The ***microfluidic*** ***device*** of claim 54, wherein said detector detects a biological response produced by said particle(s).

CLM What is claimed is:

64. The ***microfluidic*** ***device*** of claim 63, wherein said biological response is selected from the group consisting of chemotaxis, biotaxis, senescence, apoptosis, proliferation, differentiation. . .

CLM What is claimed is:

65. The ***microfluidic*** ***device*** of claim 1, further comprising a detection site, wherein said particle or product of said particle is detected by said. . .

CLM What is claimed is:

66. The ***microfluidic*** ***device*** of claim 65, wherein said detection site is within said ***microfluidic*** ***device***

CLM What is claimed is:

67. The ***microfluidic*** ***device*** of claim 65, wherein said detection site is located external to said ***microfluidic*** ***device*** .

CLM What is claimed is:

68. The ***microfluidic*** ***device*** of claim 54, wherein said detector detects a characteristic of said particle, directly or indirectly, said characteristic being selected from. . .

CLM What is claimed is:

69. The ***microfluidic*** ***device*** of claim 2, wherein said release mechanism operates by removing a retaining force caused by said retaining mechanism.

CLM What is claimed is:

70. The ***microfluidic*** ***device*** of claim 2, wherein said release mechanism operates by overcoming a retaining force caused by said retaining mechanism.

CLM What is claimed is:

71. The ***microfluidic*** ***device*** of claim 2, wherein said release mechanism operates by rendering ineffective a retaining force caused by said retaining mechanism.

CLM What is claimed is:

72. The ***microfluidic*** ***device*** of claim 2, further comprising directing said particle to another region within or external said ***microfluidic*** ***device*** .

CLM What is claimed is:

73. The ***microfluidic*** ***device*** of claim 72, wherein said another region is selected from the group consisting of a second positioning mechanism, a second. . . .

CLM What is claimed is:

74. The ***microfluidic*** ***device*** of claim 73, wherein said second retention mechanism is a cell culture chamber.

CLM What is claimed is:

75. The ***microfluidic*** ***device*** of claim 3, further comprising said output mechanism outputting said particle to a location selected from the group consisting of. . . .

CLM What is claimed is:

76. A method for perfusing cells with a reagent comprising the steps of: (a) providing a ***microfluidic*** ***device*** having (i) a cell growth chamber, a cell inlet in communication with said chamber, said cell inlet having an in. . . .

CLM What is claimed is:

77. A method for treating a particle comprising the steps of: (i) providing a ***microfluidic*** ***device*** comprising: (a) an input mechanism for introducing a fluid sample containing a particle, (b) a ***microfluidic*** passage in fluid communication with said input mechanism, (c) a positioning mechanism in fluid communication with said ***microfluidic*** passage, said positioning mechanism for positioning said particle in said ***microfluidic*** passage while contained in said fluid sample, (d) a retention mechanism for retaining said particle upon being positioned by said. . . .

CLM What is claimed is:

78. The method of claim 77 wherein said ***microfluidic*** ***device*** further comprises a release mechanism for releasing said particle from said retention mechanism, and said method further comprises the step. . . .

CLM What is claimed is:

79. The method of claim 78, wherein said ***microfluidic*** ***device*** further comprises an output mechanism for outputting said particle from said ***microfluidic*** ***device***, and said method further comprises the step of outputting said particle from said ***microfluidic*** ***device*** by said output mechanism.

CLM What is claimed is:

80. The method of claim 78, wherein said ***microfluidic*** ***device*** further comprises a cell culture mechanism for culturing said particle, and the method further comprises the step of culturing said

CLM What is claimed is:

81. The method of claim 77, wherein said ***microfluidic*** ***device*** further comprises a control mechanism for determining aspects of the flow rate or path of the sample fluid or other. . . .

CLM What is claimed is:

82. The method of claim 81, wherein said control mechanism is a valve in communication with said ***microfluidic*** passage, and the method further comprises valving said sample fluid or other fluid with said valve.

CLM What is claimed is:

83. The ***microfluidic*** ***device*** of claim 82, wherein said ***microfluidic*** ***device*** is formed from a multi-layer elastomeric block and, wherein said valve is formed from an elastomeric membrane within said elastomeric block, and wherein said valving occurs by deflecting said elastomeric membrane into said ***microfluidic*** passage.

CLM What is claimed is:

84. The method of claim 82, wherein said control mechanism is a pump in communication with said ***microfluidic*** passage, and wherein said determining the flow rate or path of said sample fluid occurs by actuation of said pump.

CLM What is claimed is:

85. The method of claim 84, wherein said ***microfluidic*** ***device*** is formed from a multi-layer elastomeric block and, wherein said pump is formed from an elastomeric membrane within said elastomeric block, and wherein said pump is actuated by deflecting a

series of elastomeric membranes into said ***microfluidic*** passage in a selected sequence.

CLM What is claimed is:
86. The method of claim 77, wherein said ***microfluidic*** ***device*** comprises a multi layered elastomeric block having a control layer having an elastomeric membrane deflectable into said ***microfluidic*** passage in a fluidic layer to selectively determine the flow rate or path of a fluid in said ***microfluidic*** passage.

CLM What is claimed is:
87. The method of claim 77, wherein said ***microfluidic*** passage further comprises an adjacent passage joining said ***microfluidic*** passage at a junction or branch, said adjacent passage being selected from the group consisting of inlet passage, outlet passage,. . .

CLM What is claimed is:
96. The method of claim 77, wherein the input mechanism is a receptacle or well in fluid communication with said ***microfluidic*** passage, and said method further comprises the step of introducing said fluid sample into said receptacle.

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=> d hist
     (FILE 'HOME' ENTERED AT 17:51:25 ON 26 MAY 2008)
     INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, ANTE, AQUALINE,
     AQUASCI, BIOENG, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS,
     CEABA-VTB, CIN, CONFSCI, CROPB, CROPU, DDFB, DDFU, DGENE, DISSABS, DRUGB,
     DRUGMONOG2, DRUGU, EMBAL, EMBASE, ...' ENTERED AT 17:52:32 ON 26 MAY 2008
                SEA MICROFLUIDIC AND RED BLOOD CELLS
              8    FILE ANABSTR
              1    FILE ANTE
              8    FILE BIOENG
             28    FILE BIOSIS
              1    FILE BIOTECHABS
              1    FILE BIOTECHDS
              1    FILE BIOTECHNO
              2    FILE CABA
             57    FILE CAPLUS
              1    FILE CEABA-VTB
              7    FILE DISSABS
             21    FILE EMBASE
             25    FILE ESBIOBASE
             65    FILE IFIPAT
              8    FILE LIFESCI
             27    FILE MEDLINE
              1    FILE NTIS
             15    FILE PASCAL
              1    FILE PHIN
              8    FILE PROMT
             66    FILE SCISEARCH
              4    FILE TOXCENTER
            580    FILE USPATFULL
            112    FILE USPAT2
             25    FILE WPIDS
             25    FILE WPINDEX
L1              QUE MICROFLUIDIC AND RED BLOOD CELLS
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             0*    FILE ANTE
             0*    FILE AQUALINE
             0*    FILE BIOENG
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             0*    FILE BIOTECHDS
             0*    FILE BIOTECHNO
             2     FILE CAPLUS
             0*    FILE CEABA-VTB
             0*    FILE CIN
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             0*   FILE FOMAD
             0*   FILE FOREGE
             0*   FILE FROSTI
             0*   FILE FSTA
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             0*   FILE KOSMET
             1    FILE MEDLINE
             0*   FILE NTIS
             0*   FILE NUTRACEUT
             0*   FILE PASCAL
             0*   FILE PHARMAML
           255    FILE USPATFULL
            48    FILE USPAT2
             0*   FILE WATER
             3    FILE WPIDS
             3    FILE WPINDEX
L2               QUE CELL(P) SIZE AND CELL(P) BIND? AND L1
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     26 MAY 2008
L3          325 S L2
L4          259 DUP REM L3 (66 DUPLICATES REMOVED)
L5          163 S L4 AND ENRICH?
L6            0 S L5 AND HIST?
L7          163 S L5 AND BIND?
L8           70 S L7 AND OBSTACLE?
L9            0 S L8 AND BINDING TO OBSTACLE?
L10          19 S L8 AND MICROFLUIDIC DEVICE

=> s l10 and cell binding
L11           5 L10 AND CELL BINDING

=> d l11 1-5

L11  ANSWER 1 OF 5  IFIPAT  COPYRIGHT 2008 IFI on STN
AN      11614553  IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI      ***MICROFLUIDIC***     ***DEVICE***   FOR CELL SEPARATION AND USES
        THEREOF
IN      Kapur Ravi; Toner Mehmet; Truskey George
PA      General Hospital Corp The (10301)
PI      US 2007264675    A1   20071115
AI      US 2007-800840        20070508
RLI     US 2005-529453        20051219 CONTINUATION             PENDING
PRAI    US 2002-414065P       20020927 (Provisional)
        US 2002-414102P       20020927 (Provisional)
        US 2002-414258P       20020927 (Provisional)
FI      US 2007264675         20071115
DT      Utility; Patent Application - First Publication
FS      CHEMICAL
        APPLICATION
ED      Entered STN: 16 Nov 2007
        Last Updated on STN: 13 Dec 2007
CLMN    27
GI      25 Figure(s).
     FIG. 1 is a schematic layout of a   ***microfluidic***     ***device***
      that enables selective lysis of cells.
     FIG. 2 is an illustration of the channel layout for the introduction of
      three fluids to the device, e.g., blood sample, lysis buffer, and
      diluent.
     FIG. 3 is an illustration of a repeating unit of the reaction chamber of
      the device where a sample of cells is passively mixed with a lysis
      buffer. In one example, 133 units are connected to form the reaction
      chamber.
     FIG. 4 is an illustration of the outlet channels of the device.
     FIG. 5 is an illustration of a device for   ***cell***   lysis.
     FIGS. 6A and 6B are illustrations of a method for the fabrication of a
      device of the invention.
     FIG. 7 is a schematic diagram of a   ***cell***     ***binding***
      device.
     FIG. 8 is an exploded view of a   ***cell***     ***binding***   device.
```

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.
FIG. 10 is an illustration of types of ***obstacles***.
FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.
FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.
FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.
FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.
FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius r
can2=0.329l. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 98%.
FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacles*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and the number of cells spiked into the sample.
FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

L11 ANSWER 3 OF 5 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11609304 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George

PA General Hospital Corp The (10301)
PI US 2007259424 A1 20071108
AI US 2007-726231 20070321
RLI US 2005-529453 20051219 DIVISION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
FI US 2007259424 20071108
DT Utility; Patent Application - First Publication
FS CHEMICAL
APPLICATION
ED Entered STN: 14 Nov 2007
Last Updated on STN: 13 Dec 2007
CLMN 34
GI 25 Figure(s)

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.

FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.

FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.

FIG. 4 is an illustration of the outlet channels of the device.

FIG. 5 is an illustration of a device for ***cell*** lysis.

FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.

FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.

FIG. 8 is an exploded view of a ***cell*** ***binding*** device.

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.

FIG. 10 is an illustration of types of ***obstacles***.

FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.

FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.

FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.3391. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of [illegible]%.

FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.

FIG. 17 is an illustration of various views of the inlet and outlets of a

cell ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

L11 ANSWER 3 OF 5 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11581736 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 20070231851 A1 20071004
AI US 2007-726276 20070321
RLI US 2005-529453 20051219 CONTINUATION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
FI US 20070231851 20071004
DT Utility; Patent Application - First Publication
FS CHEMICAL APPLICATION
ED Entered STN: 8 Oct 2007
Last Updated on STN: 9 Nov 2007
CLMN 33
GI 25 Figure(s).
FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.
FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.
FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.
FIG. 4 is an illustration of the outlet channels of the device.
FIG. 5 is an illustration of a device for ***cell*** lysis.
FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.
FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.
FIG. 8 is an exploded view of a ***cell*** ***binding*** device.
FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.
FIG. 10 is an illustration of types of ***obstacles***.
FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.
FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic

representation of the calculation of the hydrodynamic efficiency for a diagonal array. FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.
FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.
FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap2=0.329 l. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 99%.
FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu.m diameter ***obstacle*** geometry with a 50 mu.m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 mL/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.
FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.
Figures are not necessarily to scale.

AN 11523821 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA General Hospital Corp The (10301)
PI US 2007172903 A1 20070726
AI US 2007-726230 20070321
RLI US 2005-529453 20051219 DIVISION PENDING
PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
FI US 2007172903 20070726
DT Utility; Patent Application - First Publication
FS CHEMICAL

APPLICATION
ED Entered STN: 26 Jul 2007
Last Updated on STN: 16 Aug 2007
CLMN 44
GI 25 Figure(s)

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.
FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.
FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.
FIG. 4 is an illustration of the outlet channels of the device.
FIG. 5 is an illustration of a device for ***cell*** lysis.
FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.
FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.
FIG. 8 is an exploded view of a ***cell*** ***binding*** device.
FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.
FIG. 10 is an illustration of types of ***obstacles***.
FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%. FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.
FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array. FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.
FIGS. 13A-13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.
FIG. 14A is a graph of the overall efficiency as a function of pressure drop. FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.
FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rcap?=0.329l. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 98%.
FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu m diameter ***obstacle*** geometry with a 50 mu m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 mL/hr. FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.
FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series. FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device

that enables recovery of bound cells.

FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention. FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.

FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.

FIG. 24A is an illustration of a device for ***size*** based separation. FIG. 24B is an electron micrograph of a device for ***size*** based separation.

FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

L11 ANSWER 5 OF 5 IFIPAT COPYRIGHT 2008 IFI on STN

AN 11185565 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>

TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF

IN Kapur Ravi; Toner Mehmet; Truskey George

PA Unassigned Or Assigned To Individual (68000)

PPA General Hospital Corp, The (Probable)

PI US 2006134599 A1 20060622

AI US 2003-529453 20030929
WO 2003-US30965 20030929
20051219 PCT 371 date
20051219 PCT 102(e) date

PRAI US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)

FI US 2006134599 20060622

DT Utility; Patent Application - First Publication

FS CHEMICAL APPLICATION

ED Entered STN: 24 Jun 2006
Last Updated on STN: 24 Jun 2006

CLMN 69

GI 32 Figure(s).

FIG. 1 is a schematic layout of a ***microfluidic*** ***device*** that enables selective lysis of cells.

FIG. 2 is an illustration of the channel layout for the introduction of three fluids to the device, e.g., blood sample, lysis buffer, and diluent.

FIG. 3 is an illustration of a repeating unit of the reaction chamber of the device where a sample of cells is passively mixed with a lysis buffer. In one example, 133 units are connected to form the reaction chamber.

FIG. 4 is an illustration of the outlet channels of the device.

FIG. 5 is an illustration of a device for ***cell*** lysis.

FIGS. 6A and 6B are illustrations of a method for the fabrication of a device of the invention.

FIG. 7 is a schematic diagram of a ***cell*** ***binding*** device.

FIG. 8 is an exploded view of a ***cell*** ***binding*** device.

FIG. 9 is an illustration of ***obstacles*** in a ***cell*** ***binding*** device.

FIG. 10 is an illustration of types of ***obstacles***.

FIG. 11A is a schematic representation of a square array of ***obstacles***. The square array has a capture efficiency of 40%.

FIG. 11B is a schematic representation of an equilateral triangle array of ***obstacles***. The equilateral triangle array has a capture efficiency of 56%.

FIG. 12A is a schematic representation of the calculation of the hydrodynamic efficiency for a square array.

FIG. 12B is a schematic representation of the calculation of the hydrodynamic efficiency for a diagonal array.

FIGS. 13A, 13B are graphs of the hydrodynamic (13A) and overall efficiency (13B) for square array and triangular array for a pressure drop of 150 Pa/m. This pressure drop corresponds to a flow rate of 0.75 mL/hr in the planar geometry.

FIG. 14A is a graph of the overall efficiency as a function of pressure

drop.
FIG. 14B is a graph of the effect of the ***obstacle*** separation on the average velocity.
FIG. 15 is a schematic representation of the arrangement of ***obstacles*** for higher efficiency capture for an equilateral triangular array of ***obstacles*** in a staggered array. The capture radius rc=r2-0.3281. The ***obstacles*** are numbered such that the first number refers to the triangle number and the second number refers to the triangle vertex. The staggered array has a capture efficiency of 99%.
FIG. 16A is a graph of the percent capture of cells as a function of the flow rate for a 100 mu.m diameter ***obstacle*** geometry with a 50 mu.m edge to edge spacing. The operating flow regime was established across multiple ***cell*** types: cancer cells, normal connective tissue cells, and maternal and fetal samples. An optimal working flow regime is at 2.5 ml/hr.
FIG. 16B is a graph of the percent capture of cells as a function of the ratio of targets cells to white blood cells. The model system was generated by spiking defined number of either cancer cells, normal connective tissue cells, or cells from cord blood into defined number of cells from buffy coat of adult blood. The ratio of the contaminating cells to target cells was incrementally increased 5 log with as few as 10 target cells in the mixture. Yield was computed as the difference between number of spiked target cells captured on posts and number of cells spiked into the sample.
FIG. 17 is an illustration of various views of the inlet and outlets of a ***cell*** ***binding*** device.
FIG. 18 is an illustration of a method of fabricating a ***cell*** ***binding*** device.
FIG. 19 is an illustration of a mixture of cells flowing through a ***cell*** ***binding*** device.
FIG. 20A is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in series.
FIG. 20B is an illustration of a ***cell*** ***binding*** device for trapping different types of cells in parallel.
FIG. 21 is an illustration of a ***cell*** ***binding*** device that enables recovery of bound cells.
FIG. 22A is an optical micrograph of fetal ***red*** ***blood*** ***cells*** adhered to an ***obstacle*** of the invention.
FIG. 22B is a fluorescent micrograph showing the results of a FISH analysis of a fetal red blood ***cell*** attached to an ***obstacle*** of the invention. FIG. 22C is a close up micrograph of FIG. 22B showing the individual hybridization results for the fetal red blood ***cell***.
FIG. 23 is an illustration of a ***cell*** ***binding*** device in which beads trapped in a hydrogel are used to capture cells.
FIG. 24A is an illustration of a device for ***size*** based separation.
FIG. 24B is an electron micrograph of a device for ***size*** based separation.
FIG. 25 is a schematic representation of a device of the invention for isolating and analyzing fetal ***red*** ***blood*** ***cells***.

=> s L10 and device
L12 19 L10 AND DEVICE

=> s L12 and device comprising obstacle?
L13 1 L12 AND DEVICE COMPRISING OBSTACLE?

=> d

L13 ANSWER 1 OF 1 IFIPAT COPYRIGHT 2008 IFI on STN
AN 11185565 IFIPAT;IFIUDB;IFICDB <<LOGINID::20080526>>
TI ***MICROFLUIDIC*** ***DEVICE*** FOR CELL SEPARATION AND USES THEREOF
IN Kapur Ravi; Toner Mehmet; Truskey George
PA Unassigned Or Assigned To Individual (68000)
PPA General Hospital Corp The (Probable)
PI US 2006134599 A1 20060622
AI US 2003-529453 20030929

WO 2003-US30965 20030929
20051219 PCT 371 date
20051219 PCT 102(e) date
US 2002-414065P 20020927 (Provisional)
US 2002-414102P 20020927 (Provisional)
US 2002-414258P 20020927 (Provisional)
Utility; Patent Application - First Publication
CHEMICAL
APPLICATION
Entered STN: 24 Jun 2006
Last Updated on STN: 24 Jun 2006
... ***microfluidic*** ***device***
... to the ***device*** , e.g., blood sample, lysis buffer,
... with a
... one example, 133 units are connected to form the
... lysis.
... of a method for the fabrication of a
... diagram of a ***cell*** ***binding***
... view of a ***cell*** ***binding***
... ***obstacles*** in a ***cell***
... array of
... The equilateral triangle array has a capture
... calculation of the
... calculation of the
... efficiency
... pressure drop corresponds to a flow rate of 0.75 mL/hr in the
... 14A is a graph of the overall efficiency as a function of pressure
... the effect of the ***obstacle*** separation on
... capture
... the triangle vertex. The staggered array has a capture efficiency of
... 50
... and maternal and fetal samples. An optimal working flow
... of the
... number of
... between
number of spiked target cells captured on posts and number of cells

```
     spiked into the sample.
     FIG. 17 is an illustration of various views of the inlet and outlets of a
       ***cell***    ***binding***    ***device***  .
     FIG. 18 is an illustration of a method of fabricating a   ***cell***
       ***binding***    ***device***  .
     FIG. 19 is an illustration of a mixture of cells flowing through a
       ***cell***    ***binding***    ***device***  .
     FIG. 20A is an illustration of a   ***cell***    ***binding***
       ***device***   for trapping different types of cells in series.
     FIG. 20B is an illustration of a   ***cell***    ***binding***
       ***device***   for trapping different types of cells in parallel.
     FIG. 21 is an illustration of a   ***cell***    ***binding***
       ***device***   that enables recovery of bound cells.
     FIG. 22A is an optical micrograph of fetal   ***red***     ***blood***
       ***cells***   adhered to an   ***obstacle***   of the invention.
     FIG. 22B is a fluorescent micrograph showing the results of a FISH
      analysis of a fetal red blood   ***cell***   attached to an
       ***obstacle***   of the invention. FIG. 22C is a close up micrograph of
      FIG. 22B showing the individual hybridization results for the fetal red
      blood   ***cell***  .
     FIG. 23 is an illustration of a   ***cell***    ***binding***
       ***device***   in which beads trapped in a hydrogel are used to capture
      cells.
     FIG. 24A is an illustration of a   ***device***   for   ***size***   based
      separation.
     FIG. 24B is an electron micrograph of a   ***device***   for   ***size***
      based separation.
     FIG. 25 is a schematic representation of a   ***device***   of the
      invention for isolating and analyzing fetal   ***red***     ***blood***
       ***cells***  .

=> s l10 and array
L14           18 L10 AND ARRAY

=> s l14 and separation of cell?
L15            8 L14 AND SEPARATION OF CELL?

=> d l15 1-8

L15  ANSWER 1 OF 8  USPATFULL on STN
AN       2008:76951  USPATFULL <<LOGINID::20080526>>
TI       DC dielectrophoresis  ***microfluidic***   apparatus, and applications
         of same
IN       Li, Dongqing, Antioch, TN, UNITED STATES
PA       Vanderbilt University, Nashville, TN, UNITED STATES (U.S. corporation)
PI       US 2008067068           A1  20080320
AI       US 2006-523782          A1  20060919 (11)
DT       Utility
FS       APPLICATION
LN.CNT   2209
INCL     INCLM: 204/451.000
         INCLS: 204/601.000
NCL      NCLM:  204/451.000
         NCLS:  204/601.000
IC       IPCI   C07K0001-26 [I,A]; C07K0001-00 [I,C*]; G01N0027-00 [I,A]
         IPCR   C07K0001-00 [I,C]; C07K0001-26 [I,A]; G01N0027-00 [I,C];
                G01N0027-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15  ANSWER 2 OF 8  USPATFULL on STN
AN       2007:68534  USPATFULL <<LOGINID::20080526>>
TI       System for size based separation and analysis
IN       Kapur, Ravi, Stoughton, MA, UNITED STATES
         Toner, Mehmet, Wellesley, MT, MA, UNITED STATES
         Huang, Lotien R., Brookline, MA, UNITED STATES
         Barber, Tom, Cambridge, MA, UNITED STATES
         Carvalho, Bruce, Watertown, MA, UNITED STATES
         Gray, Darren, Brookline, MA, UNITED STATES
PI       US 2007059781           A1  20070315
AI       US 2005-229336          A1  20050915 (11)
DT       Utility
```

FS APPLICATION
LN.CNT 2263
INCL INCLM: 435/007.210
INCLS: 435/287.200; 702/019.000
NCL NCLM: 435/007.210
NCLS: 435/287.200; 702/019.000
IC IPCI G01N0033-567 [I,A]; G06F0019-00 [I,A]; C12M0003-00 [I,A]
IPCR G01N0033-567 [I,C]; G01N0033-567 [I,A]; C12M0003-00 [I,C];
C12M0003-00 [I,A]; G06F0019-00 [I,C]; G06F0019-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15 ANSWER 3 OF 8 USPATFULL on STN
AN 2007:68527 USPATFULL <<LOGINID::20080526>>
TI Kits for Prenatal Testing
IN Grisham, Michael, Richmond, VA, UNITED STATES
Kapur, Ravi, Stoughton, MA, UNITED STATES
Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
PI US 2007059774 A1 20070315
AI US 2005-229037 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2316
INCL INCLM: 435/007.200
NCL NCLM: 435/007.200
IC IPCI G01N0033-567 [I,A]; G01N0033-53 [I,A]
IPCR G01N0033-567 [I,C]; G01N0033-567 [I,A]; G01N0033-53 [I,C];
G01N0033-53 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15 ANSWER 4 OF 8 USPATFULL on STN
AN 2007:68473 USPATFULL <<LOGINID::20080526>>
TI Business methods for prenatal Diagnosis
IN Grisham, Michael, Richmond, VA, UNITED STATES
Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
Tompkins, Ron, Boston, MA, UNITED STATES
Schmidt, Martin, Reading, MA, UNITED STATES
Kapur, Ravi, Stoughton, MA, UNITED STATES
PI US 2007059719 A1 20070315
AI US 2005-229332 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2256
INCL INCLM: 435/006.000
INCLS: 705/002.000
NCL NCLM: 435/006.000
NCLS: 705/002.000
IC IPCI C12Q0001-68 [I,A]; G06Q0050-00 [I,A]
IPCR C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; G06Q0050-00 [I,C];
G06Q0050-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15 ANSWER 5 OF 8 USPATFULL on STN
AN 2007:68472 USPATFULL <<LOGINID::20080526>>
TI Systems and methods for ***enrichment*** of analytes
IN Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
Kapur, Ravi, Stoughton, MA, UNITED STATES
PI US 2007059718 A1 20070315
AI US 2005-229328 A1 20050915 (11)
DT Utility
FS APPLICATION
LN.CNT 2286
INCL INCLM: 435/006.000
INCLS: 435/069.100
NCL NCLM: 435/006.000
NCLS: 435/069.100
IC IPCI C12Q0001-68 [I,A]; C12P0021-06 [I,A]
IPCR C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; C12P0021-06 [I,C];
C12P0021-06 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15 ANSWER 6 OF 8 USPATFULL on STN
AN 2007:68470 USPATFULL <<LOGINID::20080526>>

```
TI      Methods for detecting fetal abnormality
IN      Balis, Ulysses, Peabody, MA, UNITED STATES
        Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
        Kapur, Ravi, Stoughton, MA, UNITED STATES
        Walsh, John, Auburndale, MA, UNITED STATES
PI      US 2007059716       A1   20070315
AI      US 2005-228454      A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2200
INCL    INCLM: 435/006.000
        INCLS: 702/020.000
NCL     NCLM:  435/006.000
        NCLS:  702/020.000
IC      IPCI   C12Q0001-68 [I,A]; G06F0019-00 [I,A]
        IPCR   C12Q0001-68 [I,C]; C12Q0001-68 [I,A]; G06F0019-00 [I,C];
               G06F0019-00 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15  ANSWER 7 OF 8  USPATFULL on STN
AN      2007:68437  USPATFULL <<LOGINID::20080526>>
TI      Veterinary diagnostic system
IN      Barber, Tom, Cambridge, MA, UNITED STATES
        Huang, Lotien R., Brookline, MA, UNITED STATES
        Gray, Darren, Brookline, MA, UNITED STATES
        Kapur, Ravi, Stoughton, MA, UNITED STATES
PI      US 2007059683       A1   20070315
AI      US 2005-229359      A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2231
INCL    INCLM: 435/005.000
        INCLS: 435/006.000; 435/007.200; 977/902.000; 977/924.000
NCL     NCLM:  435/005.000
        NCLS:  435/006.000; 435/007.200; 977/902.000; 977/924.000
IC      IPCI   C12Q0001-70 [I,A]; C12Q0001-68 [I,A]; G01N0033-567 [I,A];
               G01N0033-53 [I,A]
        IPCR   C12Q0001-70 [I,C]; C12Q0001-70 [I,A]; C12Q0001-68 [I,C];
               C12Q0001-68 [I,A]; G01N0033-53 [I,C]; G01N0033-53 [I,A];
               G01N0033-567 [I,C]; G01N0033-567 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

L15  ANSWER 8 OF 8  USPATFULL on STN
AN      2007:68434  USPATFULL <<LOGINID::20080526>>
TI      System for cell ***enrichment***
IN      Kapur, Ravi, Stoughton, MA, UNITED STATES
        Toner, Mehmet, Wellesley Hills, MA, UNITED STATES
        Huang, Lotien R., Brookline, MA, UNITED STATES
PI      US 2007059680       A1   20070315
AI      US 2005-228462      A1   20050915 (11)
DT      Utility
FS      APPLICATION
LN.CNT  2256
INCL    INCLM: 435/004.000
        INCLS: 435/006.000; 435/287.100; 435/006.000
NCL     NCLM:  435/004.000
        NCLS:  435/006.000; 435/006.000; 435/287.100
IC      IPCI   C12Q0001-00 [I,A]; C12Q0001-70 [I,A]; C12Q0001-68 [I,A];
               C12M0003-00 [I,A]
        IPCR   C12Q0001-00 [I,C]; C12Q0001-00 [I,A]; C12M0003-00 [I,C];
               C12M0003-00 [I,A]; C12Q0001-68 [I,C]; C12Q0001-68 [I,A];
               C12Q0001-70 [I,C]; C12Q0001-70 [I,A]
CAS INDEXING IS AVAILABLE FOR THIS PATENT.

=> d hist
     (FILE 'HOME' ENTERED AT 17:51:25 ON 26 MAY 2008)
     INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, ANTE, AQUALINE,
     AQUASCI, BIOENG, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CAPLUS,
     CEABA-VTB, CIN, CONFSCI, CROPB, CROPU, DDFB, DDFU, DGENE, DISSABS, DRUGB,
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     DRUGMONOG2, DRUGU, EMBAL, EMBASE, ...' ENTERED AT 17:52:32 ON 26 MAY 2008
               SEA MICROFLUIDIC AND RED BLOOD CELLS
                   9    FILE ANABSTR
                   1    FILE ANTE
                   8    FILE BIOENG
                  28    FILE BIOSIS
                   1    FILE BIOTECHABS
                   1    FILE BIOTECHDS
                   1    FILE BIOTECHNO
                   3    FILE CABA
                  57    FILE CAPLUS
                   1    FILE CEABA-VTB
                   7    FILE DISSABS
                  21    FILE EMBASE
                  25    FILE ESBIOBASE
                  65    FILE IFIPAT
                   8    FILE LIFESCI
                  27    FILE MEDLINE
                   1    FILE NTIS
                  15    FILE PASCAL
                   1    FILE PHIN
                   8    FILE PROMT
                  66    FILE SCISEARCH
                   4    FILE TOXCENTER
                 580    FILE USPATFULL
                 113    FILE USPAT2
                  25    FILE WPIDS
                  25    FILE WPINDEX
L1             QUE MICROFLUIDIC AND RED BLOOD CELLS

               SEA CELL(P)SIZE AND CELL(P)BIND? AND L1
                   0*   FILE ADISNEWS
                   0*   FILE ANTE
                   0*   FILE AQUALINE
                   0*   FILE BIOENG
                   0*   FILE BIOTECHABS
                   0*   FILE BIOTECHDS
                   0*   FILE BIOTECHNO
                   3    FILE CAPLUS
                   0*   FILE CEABA-VTB
                   0*   FILE CIN
                   0*   FILE ESBIOBASE
                   0*   FILE FOMAD
                   0*   FILE FOREGE
                   0*   FILE FROSTI
                   0*   FILE FSTA
                  18    FILE IFIPAT
                   0*   FILE KOSMET
                   1    FILE MEDLINE
                   0*   FILE NTIS
                   0*   FILE NUTRACEUT
                   0*   FILE PASCAL
                   0*   FILE PHARMAML
                 255    FILE USPATFULL
                  48    FILE USPAT2
                   0*   FILE WATER
                   3    FILE WPIDS
                   3    FILE WPINDEX
L2             QUE CELL(P) SIZE AND CELL(P) BIND? AND L1
               ---------

     FILE 'CAPLUS, IFIPAT, MEDLINE, USPATFULL, USPAT2' ENTERED AT 17:56:56 ON
     26 MAY 2008
L3           325 S L2
L4           259 DUP REM L3 (66 DUPLICATES REMOVED)
L5           163 S L4 AND ENRICH?
L6             0 S L5 AND HIST?
L7           163 S L5 AND BIND?
L8            70 S L7 AND OBSTACLE?
L9             0 S L8 AND BINDING TO OBSTACLE?
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L10            19 S L9 AND MICROFLUIDIC DEVICE
L11             5 S L10 AND CELL BINDING
L12            19 S L10 AND DEVICE
L13             1 S L12 AND DEVICE COMPRISING OBSTACLE?
L14            19 S L10 AND ARRAY
L15             8 S L14 AND SEPARATION OF CELL?

=> logoff
ALL L# QUERIES AND ANSWER SETS ARE DELETED AT LOGOFF
LOGOFF? (Y)/N/HOLD:Y
COST IN U.S. DOLLARS                              SINCE FILE      TOTAL
                                                       ENTRY    SESSION
FULL ESTIMATED COST                                   125.05     130.02

STN INTERNATIONAL LOGOFF AT 18:06:43 ON 26 MAY 2008
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